The Journal of Parasitology

Volume 27 APRIL, 1941 Number 2

VARIATION IN A NEW SPECIES OF CESTODE, *RAILLIETINA (SKRJABINIA) VARIABILA*, FROM THE PRAIRIE CHICKEN IN ILLINOIS

W. Henry Leigh*

In the course of studies carried on by the Game Management Section of the Illinois Natural History Survey relative to the status of the prairie chicken in central Illinois, fourteen young and fourteen adult birds were collected for studies of parasites and diseases (Leigh, 1940).

Ten of the fourteen young prairie chickens collected during two summers were infected with cestodes of the genus *Raillietina*. Four of the infections were so intense as to occlude the lumen of the small intestine for one-half to two-thirds its length. What effect such intense infection has upon the young hosts is not known at present. It seems reasonable that the minimum effect would be a reduction in reserve strength and vitality that might make the young more susceptible to predation or secondary infections. Finding no cestodes in the adult birds seems to indicate that the prairie chicken is susceptible to this species of *Raillietina* only or largely during the first few weeks of life.

Attempts to identify these cestodes led to the conclusion that they had not been described previously. Due to the contrast in general appearance and the variation of many morphological characters in the cestodes of several hosts, it was at first thought that the collection comprised several species. However, detailed studies revealed intergrading individuals between the extreme variants that made it clear that only one mutable species was present. This, the first representative of the genus to be recorded from the prairie chicken, is described in this paper under the name *Raillietina (Skrjabinia) variabila*.

PROCEDURE

Measurements comprising Table 1 and those used in formulating the specific diagnosis were made on only those cestodes which remained

Received for publication, November 16, 1939.

* Thanks are due Dr. R. E. Yeatter of the Illinois Natural History Survey for excellent cooperation in all phases of the problem, and to Prof. H. J. Van Cleave of the University of Illinois for critical evaluation of the results of the study.

intact, possessed gravid proglottids and were in an excellent state of preservation, namely those cestodes from hosts numbered 1, 7, and 8. The remaining part of the collection was carefully studied, but since the strobilae were immature or broken up, the data were not used in preparing the specific diagnosis.

TABLE 1.—*Synoptic characters of three differing groups of cestodes from three different hosts**

	Group 1	Group 2	Group 3
Total length, mm	6 (47–84) 71	5 (129–186) 147	4 (220–272) 247
Greatest width, mm ...	11 (0.6–2.1) 1.3	5 (1.3–1.6) 1.5	4 (2.6–2.8) 2.7
Size gravid proglottids, mm	5 (0.5–1.4 by 0.3–1.1) 0.9 by 0.5	4 (1.4–2.0 by 0.5–0.7) 1.9 by 0.6	4 (1.8–2.2 by 1.3–1.6) 2.1 by 1.5
Number rostellar hooks	3 (240–260) 252	3 Approx. 250	3 (256–272) 265
Size rostellar hooks, μ ..	1st row : 17–19 2nd row : 15–16	1st row : 18 2nd row : 15	1st row : 18 2nd row : 15–16
Size cirrus pouch, μ ...	9 : 25 (84–117 by 31–51) 90 by 39	5 : 20 (108–117 by 36–51) 113 by 45	4 : 20 (141–171 by 57–72) 159 by 64
Number testes	12 : 75 (33–65) 43	4 : 30 (56–77) 65	4 : 30 (50–82) 68
Size testes, μ.....	7 : 35 (24–39) 32	5 : 30 (39–60) 50	4 : 30 (69–102) 82
Size capsule, μ	2 : 10 (60–84 by 42–57) 72 by 47	1 : 12 (69–78 by 45–72) 72 by 61	1 : 17 (63–102 by 45–75) 86 by 59
Size embryo, μ	2 : 10 (24–30 by 21–25) 26 by 22	1 : 12 (24–30 by 21–27) 27 by 23	1 : 16 (24–30 by 19–24) 27 by 22
Size embryo hooks, μ ..	7–8	7–8	7–8
Diameter scolex, μ	15 (239–479) 315	3 (253–319) 288	4 (332–478) 406
Diameter suckers, μ ...	11 : 25 (54–96) 77	6 : 16 (81–105) 93	3 : 12 (93–114) 104
Diameter rostellum, μ ..	14 (72–129) 98	9 (81–122) 98	4 (111–143) 119
Age host	4 weeks	4 weeks	8 weeks
Number cestodes in host	35 gravid 17 immature	15 gravid	7 gravid

* Because the cestodes from these three hosts differed consistently on a number of points, they were carefully studied. In the text, the points on which these groups are separable are shown to be variously combined in an intergrading or overlapping fashion in the cestodes of other hosts. In interpreting the table, the top row of figures for any square gives the number of individuals studied and the number of measurements made for any character, with the two figures separated by a colon. The middle row, or rows, give the extremes in variation and the last row the arithmetical mean for the character.

All measurements were made without excessive flattening of the structures under consideration. Measurements of onchospheres and egg capsules were made on temporary water mounts of dissected gravid proglottids under very slight pressure. Enough measurements were made on the various structures used diagnostically to be representative of the extremes of variation of those characters and to furnish a reliable mean (See Table 1).

Numbers of rostellar hooks were determined accurately from scoleces mounted so as to present an anterior view. Measurements of rostellar hooks were made on individual hooks dissected from the scolex or on satisfactorily oriented lateral views of the hooks on mounted scoleces.

THE GENUS *Raillietina*

The genus *Raillietina* Fuhrmann, 1920, comprises numerous species found in birds and mammals. Members of the genus are characterized by a scolex armed with two rows of small hooks. Genital pores open unilaterally or alternate irregularly. The uterus breaks up into capsules containing a single or several eggs. The presence in the species here described of irregularly alternating genital pores, and egg capsules containing a single onchosphere places this form in the sub-genus *Skrjabinia*.

Previous reference to cestodes of the genus *Raillietina* in prairie chickens consists only of a single citation, that of Gross (1930) in Wisconsin. These cestodes, which were identified only to genus, in this paper are assigned to *R.* (*S.*) *variabila*. The clear differentiation of this species from described species of domestic poultry cestodes would seem to suggest that *R.* (*S.*) *variabila* is indigenous to the prairie chicken.

Raillietina (*Skrjabinia*) *variabila* n. sp.
(Figs. 1–18)

DAVAINEIDAE Fuhrmann, 1907. *Raillietina* (*Skrjabinia*) Fuhrmann, 1920. Possessing characters of the subgenus. Total length of the gravid strobilae 47–272 mm. Greatest width 0.6–2.8 mm. Final gravid proglottids measure 0.5–2.2 mm in length and 0.3–1.16 mm in width. The scolex is 239–479 μ in diameter. Suckers are 81–138 μ in length, 54–114 μ in diameter and are armed with 17–20 rows of small hooks, 8–10 μ in length. The rostellum is 72–133 μ in diameter and bears a crown of 240–272 hammer-shaped hooks, arranged in two rows. Those of the first row are 17–19 μ in length; hooks of the second row measure 15–16 μ in length. The oval to spherical testes number 33–82 in sexually mature proglottids of gravid strobilae and measure 24–102 μ in diameter. They may be small and scattered (Fig. 6) or large and closely packed together (Fig. 9) within the confines of the longitudinal excretory vessels. The vas deferens, exhibiting various degrees of convolution (Figs. 5–8), leads from the anterior poral side of the ovary to a muscular cirrus pouch which measures 84–213 μ by 33–119 μ and may extend from one half (Fig. 6) to all the way (Fig. 5) to the longitudinal excretory vessel. The extended cirrus (Fig. 10) is armed with minute spines and measures 33–66 μ by 10–15 μ. The genital pore varies in position from a point just anterior to the middle of the lateral margin of the proglottid to the anterior third of the margin (Figs. 6 and 9). The ejaculatory duct, usually visible as a slightly coiled tube within the cirrus pouch, in the cestodes of host 7 is club-shaped (Fig. 7). The ovary consists of two lateral wing-like extensions, deeply lobated in most individuals, and exhibits considerable variation in size within the mature proglottids in various hosts. The irregularly shaped vitelline gland usually lies in the region behind the posterior median indentation of the ovary, but in the Wisconsin specimen, mentioned later, appears behind the aporal wing of the ovary (Fig. 8). The distal portion of the vagina is dilated and has thickened walls (Figs. 13–14). The uterus breaks up into capsules each containing a single onchosphere. Capsules dissected from fixed gravid proglottids measured 60–102 μ by 42–75 μ. Onchospheres measure 24–30 μ by 19–27 μ (Fig. 15) and possess three pairs of hooks measuring 7–8 μ in length (Fig. 16).

Habitat: Small intestine of *Tympanuchus americanus.*
Locality: Jasper and Richland Counties, Illinois.
Date: Summers of 1936 and 1937.
Types: Collection of the Illinois Natural History Survey.

R. (*S.*) *variabila,* even with its great range of variation, is easily distinguished from the other 16 known species of the subgenus. It resembles most closely *R.* (*S.*) *centrocerci* Simon, 1937, from the sage grouse in Wyoming. Through the kindness of Mr. Simon, material of *R.* (*S.*) *centrocerci* was made available to the author for purposes of direct comparison. Simon (1937) states that *R. centrocerci* has 198–205 rostellar hooks. This range was verified on four anterior views of mounted scoleces. Testes were said to number 63–118. Counts made on three strobilae of Simon's material gave a range of 111–158 testes per sexually mature proglottid. In view of its smaller size, more numerous rostellar hooks and significantly smaller number of testes it is believed that *R. variabila* constitutes a separate species.

It should be pointed out that Simon has erroneously included *R.* (*Raillietina*) *joyeuxi* in the subgenus *Skrjabinia.* Fuhrmann (1932) places this species in the subgenus *Raillietina.*

DISCUSSION OF VARIATION

Those studies on variation in cestodes which have been located in the literature are concerned largely with observations on abnormalities in form and structure and not with variation in normally formed individuals (Child, 1900 and 1902; Dobrovolny, C. G. and M. D., 1935), hence are not directly related to the present problem.

Because this collection of cestodes was at first believed to represent more than one species, most of the observations were made on those well-preserved variants which seemed to possess differences of more than specific magnitude. Consequently, those early measurements are preserved in Table 1. The discussion below will indicate the cospecific nature of the variants. The three groups mentioned in the discussion are groups 1, 2, and 3 of Table 1 and were from hosts 1, 7, and 8, respectively. It was these three groups that received the greatest study. Cestodes from other host individuals will be mentioned in the discussion as the occasion arises.

Size of Strobilae

Groups 1, 2, and 3 (Table 1) (Figs. 6, 7, 9) may be clearly separated on the basis of total length and width of complete gravid strobilae. The explanation for this appreciable difference in size of cestodes from host to host is not clear. Number of individuals and available food, age of parasite and host, and individual differences in host resistance are possible factors. It is significant that the host harboring the smallest

number of cestodes also harbored the largest. Due to size differences the 7 cestodes of host 8, the 15 cestodes of host 7, and the 52 cestodes of host 1 all filled the small intestines to an approximately equal degree.

Scolex Characters

Number and size of rostellar hooks always receive considerable attention in species determination. Descriptions of some members of this subgenus indicate little or no variation in number of rostellar hooks. *R.* (*S.*) *microcotyle* (Lopez-Neyra, 1931) is reported as having 34 hooks; *R.* (*S.*) *magnicornata* (Fuhrmann, 1909), *R.* (*S.*) *circumvallata* (Joyeux & Baer, 1936) and *R.* (*S.*) *polyuterina* (Fuhrmann, 1909) each have 200 rostellar hooks. *R.* (*S.*) *lavieri* (Joyeux & Baer, 1928) possesses 70–95; *R.* (*S.*) *ransomi* (Williams, 1931) has 500–520 and *R.* (*S.*) *cesticillus* (Joyeux & Baer, 1936) has been observed to have 200–300 in some instances and 400–500 in others. Other members of the subgenus have a range of variation from 7 to 100 hooks. Thus the literature indicates that some species of this genus may be variable with respect to number of rostellar hooks and others relatively constant. The range of 42 hooks in *R.* (*S.*) *variabila* is not unusual for this genus. Other scolex characters such as size of scolex, rostellum and suckers, and armament of the suckers offered no significantly contrasting characters within the three groups.

Number and Size of Testes

The number of testes within certain limits is considered a useful diagnostic character for members of this genus. An analysis of the number of testes for representative cestodes of the entire collection has given evidence of greater intraspecific variation than is usually encountered in cestode literature. The greatest range of variation reported for any member of this subgenus was found in *R.* (*S.*) *centrocerci* (Simon, 1937). This range was 63–118, and the author, using some of Simon's material, counted as many as 158 testes in sexually mature proglottids. *R.* (*S.*) *crepidocotyle* (Joyeux & Baer, 1935) is reported as having 50–70 testes; descriptions of the other members of the subgenus indicate no range of variation greater than 10. The entire range of variation in *R.* (*S.*) *variabila* lies between 82 and 33 testes in typical mature proglottids of gravid strobilae. Non-gravid strobilae of group 1 had as low as 17 testes in the sexually mature proglottids. A glance at Table 2 will reveal minor gradations in the maximum and minimum numbers of testes for individual strobilae. It is interesting to note that the minimum numbers of testes for some strobilae exceed the maximum numbers for other strobilae.

Table 1 shows discontinuous variation in size of testes of groups 1 to 3. In cestodes of group 1 the testes are small and scattered (Fig. 6)

TABLE 2.—*A study in the range of variation of number of testes within single complete strobilae, giving the maximum number for the immature proglottids, and maximum and minimum numbers in the mature proglottids for each strobila**

No. of ind.	Maximum no. in immature proglottid	Maximum no. in mature proglottid	Minimum no. in mature proglottid	Amount of variation between extremes
1.6	63	63	42	21
1.1	66	57	38	19
1.1	55	52	37	17
1.5	51	47	33	14
1.16 imm.	58	46	28	18
1.12 imm.	53	39	26	13
1.15 imm.	48	29	17	12
7.3	..	77	62	15
7.1	76	76	58	18
7.2	80	75	56	19
8.7	82	82	60	22
8.1	77	76	62	14
8.9	68	68	57	11
8.8	65	65	50	15
Wisc.	..	56	44	12

* The left hand column gives the number of the host and number of the cestode studied. Cestodes of hosts 1, 7, 8, representing groups 1, 2 and 3 of Table 1, were those used for this table. All specimens with the exception of those marked imm. (immature) were gravid strobilae. "Wisc." refers to the Wisconsin specimen. In some instances the minimum number of testes of cestodes in hosts 7 and 8 exceeds the maximum number for cestodes of host 1.

in contrast to the closely packed, large testes of group 3 (Fig. 9.) Without a consideration of other collections of cestodes, these two groups would undoubtedly be regarded as distinct species. Cestodes from other hosts of the same area and from a prairie chicken in Wisconsin had proglottids with 44–56 testes, the numbers characteristic of group 1, but with testes the size of those of groups 2 and 3 (Figs. 8 and 9).

Size and Extent of Cirrus Pouch

Within limits the size and the extent to which the cirrus pouch reaches medially towards the longitudinal excretory vessel have been considered as sufficiently invariable in this genus to emphasize them in description. In this study, the cirrus has been found to vary within wide limits. While its size is relatively constant within a single strobila and even in the individuals of a single host, the cirrus pouch has shown marked discontinuous variation from one host to another (Table 1).

In the cestodes of other hosts, not included in the table, cirrus and testes characters of the three groups are variously and confusingly combined. For instance, cestodes from one host had 33–42 testes, thus conforming to cestodes in group 1, but the cirrus pouch measured 170–213 μ by 87–119 μ and extended to the longitudinal excretory vessel, thereby exceeding the size of the cirrus pouch of cestodes of group 3 which are larger in length and width of strobilae. For all specimens studied, a range of 129 μ in length and 86 μ in width has been observed. A range of 50 μ in length is the greatest variation reported for any other member of the subgenus. *R.* (*S.*) *lavieri* (Joyeux & Baer, 1928) and *R.* (*S.*)

crepidocotyle (Joyeux & Baer, 1935) each exhibited this amount of variation.

The extent to which the cirrus pouch extends medially toward the longitudinal excretory vessel in sexually mature proglottids varies from a point slightly under half way to the vessel through gradations up to the vessel itself (Figs 5–9). Also in gravid proglottids, if they are narrow, the pouch may rest against the vessel (Fig. 10), whereas in wide ones, the cirrus may not reach half way to the vessel (Fig. 12). So the size and position of the cirrus pouch in this species hold limited significance as diagnostic characters.

Egg Capsule and Onchosphere

In contrast to the variation observed in the several characters discussed above, the onchospheres and egg capsules of the variants show little dissimilarity. Measurements of egg capsules and onchospheres do not change significantly in any of the groups (Table 1) and all conform to a common pattern (Fig. 16).

Atypical Cestodes

Mention should be made of a rather striking variation evident in certain strobilae of host 1, since they are sharply set off from the other members of the group. The entire strobilae are markedly smaller, and the maximum and minimum numbers of testes are below the figures for the typical members of the group. Those individuals in Table 2 marked imm. (immature) represent this group with a minimum number of 17 testes.

In these atypical individuals (Fig. 4) sexually mature proglottids are unusually narrow. The ovary remains small and undeveloped in comparison with the enlarging lobated ovary of the typical mature proglottid. Testes, in addition to being few in number, are unusually small. The final proglottids possess recognizable remains of ovary and testes, but there is no indication that the uterus is breaking up into egg capsules enclosing onchospheres. Whether these atypical members are younger and later attain the power of reproduction or for some reason are degenerate and never function reproductively can only be conjectured. Their irregularity might be associated with heavy infestation, as this four-week-old host had 52 cestodes packing the small intestine.

Young (1935), in reviewing unsolved problems of cestode structure and development, states that variation in the number of testes is related to the problem of the origin of germ cells. He had observed testes to arise from such specialized somatic tissues as muscle cells. Child (1902) observed "that reduction in size of female organs and reduction in number of testes frequently occurs in proglottids less than normal size."

Wisconsin Specimens

Unconformity in this species was further observed in fragments of cestodes taken from a prairie chicken in Wisconsin by Gross in 1930. This material was obtained from the Bureau of Animal Industry through the kindness of Dr. E. W. Price. These fragments, while obviously belonging to the subgenus *Skrjabinia,* combined characters of groups 1 and 3, the largest and smallest of the groups mentioned above. The one scolex obtained had hooks 16–18 μ long and, while they could not be accurately counted, were clearly numerically close to the Illinois specimens. The testes numbered 44–56 per proglottid, thus were within the range of 37–65 for group 1. However, they measured 60–93 μ in diameter, and this measurement conforms closely to the range as found in group 3, not at all to the range of group 1. The sexually mature proglottids were 1.82 mm in width, thus smaller than those of group 3, but the cirrus pouch exhibited higher maximum and average measurements, a maximum measurement of 135–180 μ by 68–81 μ and an average of 167 by 65 μ. Measurements of egg capsules and onchospheres could not be made. Thus, within a single strobila (Fig. 8) from a Wisconsin prairie chicken were found characters that might possibly have been used in separating groups 1 (Fig. 6) and 3 (Fig. 9) into separate species.

The above discussion presents what has seemed to the author an unusual amount of variation within a single species of cestode, as compared with descriptions of other described members of the subgenus *Skrjabinia.* The fact that all the hosts (Wisconsin specimen excepted) were collected within two counties and that the variations observed were similar for the cestodes of a single host, but dissimilar from one host to another, would seem to indicate that the explanation of this variation lies in a study of the relationships of parasites to their individual hosts.

SUMMARY

1. A new species of cestode, *Raillietina* (*Skrjabinia*) *variabila,* has been described. It is the first species of this genus described for the prairie chicken, *Tympanuchus americanus* (Reichenbach).

2. This species was found in ten of fourteen young prairie chickens collected in two adjacent Illinois counties. In four cases the infestations were so intense as to occlude the lumen of the small intestine.

3. Size of complete gravid strobila, number and size of testes, size and extent of cirrus pouch are characters which vary within wide limits in this species.

4. Number, size and shape of rostellar hooks, size and appearance of egg capsules, onchospheres and onchosphere hooks are relatively constant throughout the entire collection of cestodes.

5. *Raillietina* (*Skrjabinia*) sp. from a prairie chicken in Wisconsin collected by Gross (1930) has been assigned to *R.* (*S.*) *variabila.*

6. Variations observed in *R.* (*S.*) *variabila* were constant for specimens within individual hosts, but noticeably different within different hosts of the same area, indicating a considerable genetic tolerance of the species to variation of the several characters mentioned. Factors responsible for variation have not been determined.

Bibliography

Child, C. M. 1900 Abnormalities in the cestode *Monieza expansa*. I. Biol. Bull. **1**: 215–261.

———1902 Abnormalities in the cestode *Monieza expansa*. III. Biol. Bull. **3**: 95–114 and 143–160.

Dobrovolny, C. G. and Dobrovolny, M. D. 1935 Polyradiate tapeworms from a dog. Tr. Am. Micr. Soc. **54**: 22–27.

Fuhrmann, O. 1909 Neue Davaineiden. Central Bakt. **49**: 94–124.

———1920 Considerations generals sur le genre Davainea. Festschrift für Zschokke. No. 27: 1–19.

———1932 Les Tenias des Oiseaux. Mem. Univ. Neuchatel. **8**: 1–381.

Gross, A. O. 1930 Progress report of the Wisconsin prairie chicken investigation. Bull. Wisconsin Conservation Comm. pp. 1–112.

Joyeux, Ch. and Baer, J. G. 1928 Sur quelques cestodes de la region d'Entebbe (Uganda). Ann. Parasitol. **6**: 179–181.

———1935 Cestodes d'Indochine. Rev. Suisse Zool. **42**: 249–274.

———1936. Cestodes. Faune de France. **30**: 1–613.

Leigh, W. H. 1940 Preliminary studies on parasites of upland game birds and fur-bearing mammals. Bull. Illinois Nat. Hist. Survey **21**: 185–194.

Lopez-Neyra, C. R. 1931 Revision del genero Davainea. Mem. Acad. Ciencias Exactas, Fisicas y Naturales **1**: 1–179.

Simon, F. 1937 A new cestode, *Raillietina centrocerci*, from the sage grouse, *Centrocercus urophasianus*. Tr. Am. Micr. Soc. **56**: 340–343.

Williams, O. L. 1931 Cestodes from the eastern wild turkey. J. Parasitol. **18**: 14–20.

Young, R. T. 1935 Some unsolved problems of cestode structure and development. Tr. Am. Micr. Soc. **54**: 229–239.

Explanation of Figures

All figures drawn with the aid of a camera lucida. Figs. 1 and 2 drawn to scale of Fig. 2; Figs. 4–9 to scale of Fig. 4; Figs. 10–12 to scale of Fig. 12; Figs. 13–18 to scale of Fig. 17. Hooks of Figs. 1–3 drawn in by hand and not representative of accurate numbers.

Fig. 1. Lateral view of scolex, group 1.

Fig. 2. Anterior view of scolex, group 3.

Fig. 3. Lateral view of rostellum, showing arrangement of hooks.

Fig. 4. Atypical proglottid group 1, drawn to same scale as Figs. 5–9.

Fig. 5. Proglottid combining some features of Figs. 6 and 7.

Fig. 6. Sexually mature proglottid, group 1.

Fig. 7. Sexually mature proglottid, group 2.

Fig. 8. Sexually mature proglottid, from Wisconsin specimen.

Fig. 9. Sexually mature proglottid, group 3.

Figs. 10, 11, 12. Final gravid proglottids of groups 1, 2, and 3, respectively, drawn to same scale.

Fig. 13. Cirrus pouch-vagina complex, cirrus unextruded.

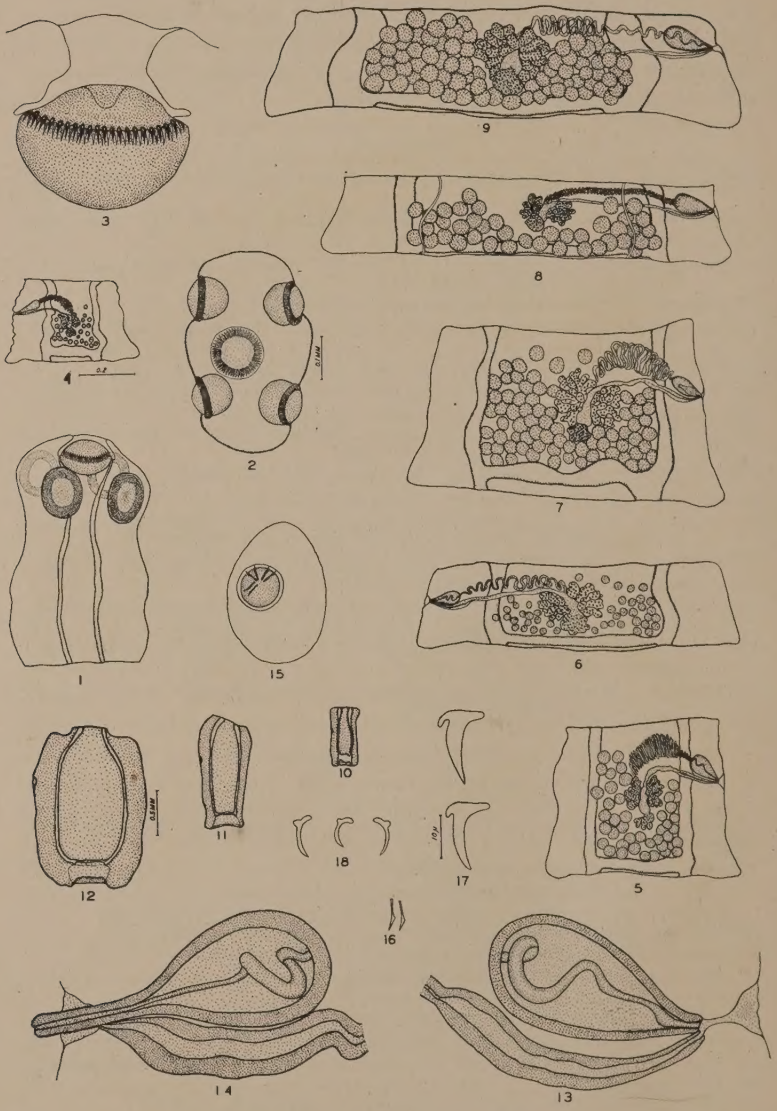
Fig. 14. Cirrus pouch-vagina complex, spined cirrus extruded.

Fig. 15. Egg capsule showing onchosphere and inner envelope.

Fig. 16. Onchosphere hooks.

Fig. 17. Rostellar hooks.

Fig. 18. Acetabular hooks.



Raillietina (Skrjabinia) variabila

RECOVERY OF *TRICHINELLA SPIRALIS* LARVAE IN EARLY STAGES OF INFECTION

ARTHUR J. LEVIN*

Department of Zoology, State University of Iowa, Iowa City, Iowa

Although the life cycle of *Trichinella spiralis* is known in a general way, many of the details remain obscure, due to the inadequate methods employed in their study. To cite one instance, the early phases of the life cycle of *Trichinella* have not been investigated because the methods available for obtaining larvae in later stages of infection have not been of use in recovering the young worms. Leuckart (1) and his contemporaries used a method of scraping and chopping the meat and washing the larvae out for recovery of worms in all stages of infection. Ransom (2) devised the method of pepsin digestion, and the workers in the field immediately took up this cleaner recovery technique. However, since the young worms cannot withstand this digestive treatment (McCoy, 3), no publications have appeared on this phase of trichina investigation since that time. This present study was undertaken for the purpose of establishing a quantitative method for the recovery of larvae in the early stages of infection.

MATERIALS AND METHOD

The rats used were albino and hooded laboratory stock, between 11 and 20 weeks old. They were fed on a diet of dog biscuits and water, and kept under the same conditions. The *Trichinella* larvae used for infection were obtained from rats, infected eight weeks or more, by the method of digestion described by Ransom. After having previously been starved for 24 hours, each group of rats was infected at the same time with a dose of 30 larvae per gram body weight introduced into the stomach with a pipette. When the predetermined time of infection had elapsed, each rat was killed and the larvae recovered as described below:

1. Strip the skin and eviscerate the rat.
2. Grind the meat through a fine mesh grinder. Do not allow the meat to become dry at any time from this point on.
3. Separate ground meat into parts of not more than 25 grams each. Larger masses necessitate many washings to recover all the larvae.
4. Place each 25 grams (or less) of meat into a 500 cc Erlenmeyer flask, and add 60 cc 0.9% saline for each 10 grams of meat. Introduce glass beads into the flask and shake vigorously for 3 to 4 minutes to further break up the muscle fibers.
5. Filter off all the liquid through a 60 mesh screen placed in a Buchner funnel in a suction flask. To keep the meat in the flask while pouring the fluid off,

Received for publication, June 4, 1940.

* The author is deeply indebted to Dr. L. O. Nolf for his invaluable guidance during the course of this investigation, and for his helpful suggestions in preparing the manuscript.

the opening of the flask should be covered with cheesecloth. Any meat adhering to the cloth can be washed back by pouring saline over the cloth into the flask. Discard each piece of cheesecloth after being used once.

6. Wash meat by adding 30 cc 0.9% saline per 10 grams meat and shake the mixture for 2 to 3 minutes. Filter off the liquid and add to the first filtrate.

7. Add 60 cc of 0.9% saline for each 10 grams of meat and connect the flask to a compressed air hose. Force air through the mixture vigorously for one hour at room temperature (22° C). Higher temperatures cause a froth to form, making it more difficult to wash all the larvae out of the meat.

8. Filter off the liquid through a 60 mesh screen and add to the first filtrate.

9. Wash meat *twice* as described under (6). Add all liquid to the first filtrate.

10. Centrifuge the total filtrate suspension to concentrate the larvae. The supernatant fluid should be examined for larvae before being discarded. The concentrated larvae can be kept indefinitely by adding some 5% formalin (5 cc) and storing at 5° C.

11. Make larvae counts by drawing 0.1 cc samples from the larvae concentrate made up to known volume (McCoy, 4) and count under the low power of the microscope on a ruled glass slide.

In working out this method, the meat was washed 10 to 15 times after being subjected to the compressed air treatment, and only in three cases were any additional larvae found after the first washing; 50 larvae were found in one case on the 14th day after infection, 10 larvae on the 16th day after infection, and 100 larvae on the 17th day after infection. The second washing was therefore added to the technique to insure recovery of all the larvae.

EXPERIMENTS AND RESULTS

Each group consisted of ten rats infected at the same time with larvae from the same source. The first rat was killed five days after infection, the second six days later, and so on through fourteen days. Two groups of rats were so treated. A third group consisted of five rats. The first was killed 5½ days after infection, the second 6 days after infection, the third 8 days after infection, the fourth on the 9th day and the fifth 11 days after infection. The infective dose consisted of 30 larvae per gram body weight administered per os. No larvae were recovered five and a half days after infection. On the 6th day, as many as 50,000 worms were recovered, ranging in size from 95 to 125 microns in length by 5 microns wide, which is the size of larvae forced from a gravid female. From the 7th to the 11th day inclusive, the increase in numbers of larvae is marked; 135,000 being recovered on the 7th day, 151,000 on the 8th day, 221,000 on the 9th day, 316,000 on the 10th day, and 350,000 on the 11th day. The number recovered thereafter remained relatively constant; 415,000 on the 12th day, 435,750 on the 13th day, and 415,000 on the 14th day. Larvae-larvae ratio calculations were made on the basis of the number of larvae fed and the number of larvae recovered each day. These results are summarized in Table 1, and graphically in Fig. 1. Daily measurements of the larvae indicate that the larvae of 14 days infection reach a maximum size of 375 microns in length and 22.5 microns in width. How-

ever, the size and growth relationship of the young larvae are being studied at present and will be summed up in another paper.

The young larvae, up to 12 days after infection, could be recovered alive when the following modification of the maceration method was used:

The entire muscle portion of the rat, finely ground, was placed in 0.9% saline in a flask (see Nos. 1 and 2 under Method) and shaken gently. Then the liquid was filtered off through a fine mesh screen (see No. 5 under Method). The filtrate contains the living larvae. This

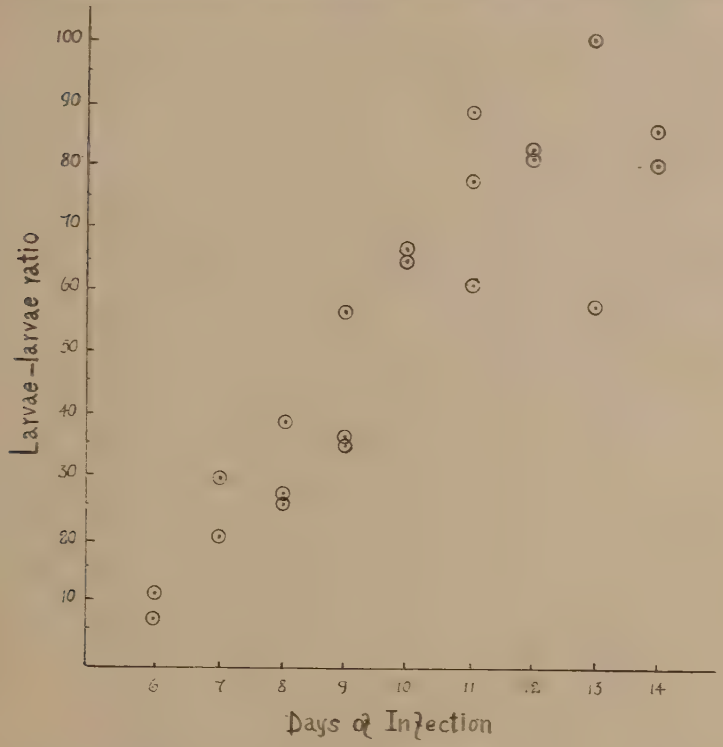


FIG. 1. A graphic representation of the daily larvae-larvae ratio (number of larvae recovered divided by the number of larvae fed).

modification of the maceration method is of use only in obtaining the young larvae alive. The number of larvae recovered in this way is in no sense a true count of the total number of larvae contained in the muscles of the rat. From the 12th day after infection on, many of the young worms will remain alive even after having been put through the complete process of maceration, as outlined under Method.

A fourth group of rats was used to find how long after infection the method could be used efficiently. One rat was killed 16 days after infec-

TABLE 1.—*Data from Groups 1, 2, and 3*

Rat	Days infected	Weight at infection, grams	No. larvae fed	Weight when killed, grams	No. larvae recovered	Larvae-larvae ratio
*I— 1	5	188	5640	143	0	...
II—11	5	187	5630	168	0	...
III—22	5½	134	4020	...	0	...
I— 2	6	174	5220	133	0	...
II—12	6	174	5200	143	32,200	6.2
III—30	6	174	4500	119	50,500	11.2
I— 3	7	188	5640	132	120,000	21.3
II—13	7	169	5060	137	149,000	29.4
I— 4	8	182	5460	131	145,000	26.6
II—14	8	184	5500	160	151,875	27.6
III—31	8	135	4050	124	155,000	38.3
I— 5	9	226	6780	159	237,500	35.0
II—15	9	161	4840	138	172,500	35.6
III—27	9	150	4500	126	253,000	56.2
I— 6	10	165	4900	102	330,000	67.3
II—16	10	161	4830	145	317,272	65.7
I— 7	11	159	4770	117	295,000	61.8
II—17	11	168	5050	144	450,000	89.1
III—36	11	150	4500	126	350,750	77.9
I— 8	12	167	5010	126	415,000	82.8
II—18	12	165	4960	143	400,000	80.6
I— 9	13	240	7200	151	414,750	57.6
II—19	13	153	4570	122	458,750	100.4
I—10	14	169	5070	151	430,250	84.9
II—20	14	169	5070	134	401,250	79.1

* The Roman numerals and Arabic numbers (e.g. II—19) refer to the numbers of individual experiments.

tion, another at 17 days, another at 21 days and the last at 28 days. As previously noted, larvae were recovered after the first washing on the 14th, 16th, and 17th days. This seemed to indicate that there was a time after which this method would be less efficient. By the 21st day, the maceration method was of no value in recovering the larvae. The digestion method (80:1 pepsin dilution) was efficient from this point on. Table 2 summarizes the results of this group.

Some rats yielded results far out of line with the majority of cases. Possible causes for these variations will be discussed later.

TABLE 2.—*Data from Group 4*

Rat	Days infected	Weight at infection, grams	No. larvae fed	Weight when killed, grams	No. larvae recovered	Larvae-larvae ratio
*IV—25	16	170	5100	173	117,000	22.9
IV—29	17	150	4500	143	152,250	33.8
IV—34	21	142	4240	140	380,750 (total) maceration: 236,250 digestion: 144,500	89.8
IV—35	28	146	4380	152	263,750** (total)	60.2

* The Roman numerals and Arabic numbers (e.g. IV—29) refer to the numbers of individual experiments.

** The ground meat from this rat was divided into two equal parts: A and B. Part A was first macerated and then digested. Part B was digested only. Part A. Maceration: 17,000 larvae recovered; digestion: 112,500 larvae recovered. Part B. Digestion: 134,250 larvae recovered.

DISCUSSION

The maceration method utilized here is based on the consideration that the young larvae can be recovered by mechanical agitation of finely ground meat because they do not become encapsulated in the muscle sheath until at least four weeks after infection.

Most observers are agreed upon the fact that the first larvae appear in the host tissue about the fifth or sixth day after infection (Leuckart (1), Virchow (5), and others). In this work, no larvae were recovered until the sixth day. The duration of larval production and larval migration has been estimated by various authors at from six to twenty-five days (Leuckart (1), et al; Augustine (6)). According to Leuckart, the greatest migration takes place on the 9th and 10th days and the maximum invasion of muscle occurs between the 12th and 15th days. Augustine places the maximum invasion on the 10th day. This present work seems to indicate that the deposition of larvae, once it is begun, is a rapid and constant process, occurring over a period of six days. Larval migration seems to be of very short duration, and penetration of the host tissue seemingly takes place very shortly after the larvae have been released. Since the number of larvae recovered after the 12th day does not increase, it seems correct from these experiments, to set the duration of larval production and migration at from the 6th to the 12th days.

The small intestine of each rat was examined for the presence of adult worms. Up to the 16th day, adults were found. On the 16th day and thereafter, no adults were found in the intestine. This corresponds exactly with the findings of Christenson (10) and Gursch (7).

Although most of the data checks with a fair degree of accuracy, variations do occur. These variations seem to be of two kinds: one, wherein the number and size of the larvae recovered on a particular day are smaller than that ordinarily found; and two, wherein the number of larvae recovered on a particular day is less than might be expected, based on previous experiments, although the size of the larvae corresponds with the size of those usually found on that day. The former type of variation will be discussed in a subsequent paper on size and growth relationships of the larvae.

The latter type of variation is exemplified particularly in Group IV. Gursch (7), working in this laboratory, using the same stock of rats, the same source of larvae and the same infection dose, found similar variations. In one case, in a 12 day infection, he could recover only 15 per cent of the original dose from the intestine, whereas 42 per cent recovery is usual. Furthermore, in another rat, examined 10 hours after infection, he found 37 per cent of the larvae still in the stomach, whereas all the other cases showed less than one per cent. According to Glazier (8), Flint states that the liberation of trichinae in the stomach corresponds

with the time required for solution by the gastric juice of the albuminous substances of the muscle, which takes from one and a half to five hours. Professor Wedl, in a report to the Medical Society of Vienna in 1867, further states that in vitro, free trichinae are acted on and killed by gastric juice, while it only serves to dissolve the capsule in other cases and thus set free the worm. From this, one might postulate that worms, previously digested out and fed, and then remaining for an additional ten hours in the stomach of the host would take little, if any part in producing an infection.

McCoy (9) was able to demonstrate that, in immunized rats, larvae fed were quickly passed in the feces. It is conceivable that this might occur in rats showing a natural immunity. Thus, the retention of larvae in the stomach, and the passing out of larvae in the feces before producing an infection could account for a low infection in non-immunized rats.

SUMMARY

1. Young *Trichinella* larvae, in infections of from six days to twenty-one days, can be recovered from rat tissue by a technique of maceration. This technique separates the larvae from host tissue by physical means, without resort to chemicals, such as pepsin, which destroy the young worms.

2. When an infective dose of approximately 5000 larvae was used, the first evidence of larvae in the rat tissue was obtained about six days after infection.

3. The numbers of larvae released into the rat tissue increase rapidly from the sixth to the eleventh day. By the twelfth day, the great majority of larvae have apparently been released. The numbers recovered remain fairly constant from the twelfth day onward.

4. The size of the larvae recovered from rat tissue on the sixth day is the same as the size of larvae squeezed from a gravid female.

5. Extreme variations from the normal (majority of cases) in the number of larvae recovered may be accounted for by the fact that, in some instances, the larvae fed may have remained in the stomach for abnormally long periods of time and died before passing into the small intestine; and the possibility of their being passed in the feces before taking part in producing an infection.

BIBLIOGRAPHY

1. LEUCKART, R. 1876. "Menschlichen Parasiten," Leipzig **2**: 358, 531, 548, 568.
2. RANSOM, B. 1916 Effect of refrigeration upon larvae of Trichinella spiralis. J. Agric. Res. **5**: 819–854.
3. MCCOY, O. R. 1934 The effect of vitamin A deficiency on the resistance of rats to infections with *Trichinella spiralis*. Am. J. Hyg. **20**: 169–180.
4. MCCOY, O. R. 1931 Immunity of rats to re-infection with *Trichinella spiralis*. Am. J. Hyg. **14**: 484–494.

5. Virchow, R. 1864 "Die Lehre von den Trichinen," Berlin, p. 34.
6. Augustine, D. 1938 "Parasitology," Hegner, Root, Augustine, Huff. pp. 476–478.
7. Gursch, O. F. 1940 Personal communication.
8. Glazier, W. C. W. 1881 "Report on Trichinae and Trichinosis." Treasury Dept. Document 84, Marine Hosp., Washington, D. C.
9. McCoy, O. R. 1938 Rapid loss of *Trichinella spiralis* larvae fed to immune rats and its bearing on the mechanism of immunity. J. Parasitol. **24** Suppl.: 35 (abstract).
10. Christenson, R. O. 1927 Sex ratio of adult trichinae. Science **66**: 259.

DISCOVERY OF HUMAN HEARTWORM INFECTION IN NEW ORLEANS*

ERNEST CARROLL FAUST, E. PERRY THOMAS, AND JACK JONES

Heartworm infection in domestic dogs is prevalent throughout Southern Europe, China, Japan, the Netherlands Indies, Fiji, New Caledonia, Australia, Hawaii, and the warmer regions of North and South America. In some of these areas the incidence in dogs over one year of age is as high as 30 to 50 per cent. The etiological agent of this infection is *Dirofilaria immitis* (Leidy, 1856). This same species of filaria has also been recorded from the dingo (*Canis dingo*) in Australia; from *C. brachyurus* in Brazil; from the wolf (*Canis* sp.) in Japan; from the timber wolf (*C. floridanus*) near New Orleans, La.; from the fox (*Vulpes vulpes*) in North China; from the domestic cat (*Felis catus domestica*) in Virginia (U. S. A.), Dutch Guiana and North China; from the tiger (*F. tigris*) in French Indo-China; from *F. tigris sondiaca* in the Netherlands Indies, and from the jaguar (*F. onca*) in Surinam. Moreover, the specimens of *Dirofilaria* obtained from the common seal (*Phoca vitulina*), the crested seal (*Stemmatopus cristatus*) and the California sea-lion (*Zalophus californianus*) are apparently indistinguishable morphologically from the dog heartworm (Faust, 1937). Another species of heartworm (*D. pongoi*) has been described by Vogel and Vogelsang (1930) from the Borneo orang-outang (*Pongo pygmaeus*), and still another species (*D. indica*) by Chakravarty (1936) from a Calcutta dog.

With the possible exception of a few observations made in Tokyo by Tokishige (cited in Blanchard, 1895), these worms have been recovered from the chambers of the right heart, the inferior and superior vena cava, and the pulmonary artery, or, less frequently, have migrated from the pulmonary artery into the respiratory tree. Tokishige reported 82 heartworm infections in dogs, of which one occurred in the left heart, one in the posterior (abdominal?) aorta, and one in the crural artery, while in one instance the worms had perforated the interauricular septum.

In 1887 de Magalhães reported the discovery of a single male and a single female filaria in the left ventricle of a male child from Rio de Janeiro. These worms were studied by Blanchard (1896), who stated

Received for publication, June 7, 1940.

* From the Departments of Tropical Medicine and Gynecology, School of Medicine, Tulane University of Louisiana, New Orleans, La. We are deeply indebted to Dr. Herman Lent of the Instituto Oswaldo Cruz, Rio de Janeiro, for providing us with a photostatic copy of the original de Magalhães paper and for the statement that the specimen described by Professor de Magalhães and later examined by Professor Blanchard has been lost, presumably in a fire which destroyed the de Magalhães home.

that de Magalhães' description was accurate but recognized that the worms were specifically different from the dog heartworm and named them *magalhãesi* in honor of their discoverer.

The heart filarias all belong to the subgenus *Dirofilaria* Faust, 1937, as distinguished from the species of *Dirofilaria* living in the cutaneous and subcutaneous tissues (subgenus *Nochtiella* Faust, 1937).

PRESENTATION OF DATA

The present report concerns a second case of human heartworm infection, from an aged negress, native and life-time resident of New Orleans, Louisiana.

The woman had never gone into the country for a visit or to work. Her only absence from the city had occurred more than twenty years ago, when she was placed in a government hospital for treatment of drug addiction. She died at the age of 73, probably as a result of acute toxic nephritis. The body did not come to autopsy but was embalmed and later was utilized for dissection. In the abdominal cavity there was a peritoneal abscess around the cecum, extending along its medial and lateral sides; it was adequately walled-off by the omentum and small intestine. There was no evidence of edema or abdominal ascites. The kidneys presented gross evidence of acute toxic nephritis. The pleura, lungs and heart showed no gross pathology.

On dissection of the inferior vena cava a stiff, creamy-white thread was discovered projecting out of a blood clot. After the object had been washed and placed in formalin it was evident that it was no artefact. Examination with the naked eye and with a hand lens suggested that it was a male filaria, especially in view of the characteristic corkscrew coiling of its more attenuated end. Search for a female worm was unsuccessful and examination of a portion of the blood clot for microfilariae was negative. It was then concluded that the infection consisted of a single male worm, which was cleared in glycerin and was found to be a specimen of *Dirofilaria*. Additional study was then undertaken to discover whether (1) it was *Dirofilaria immitis,* which is quite common in dogs in the New Orleans area, (2) *D. magalhãesi,* or (3) a new species of *Dirofilaria*.

DESCRIPTION OF THE WORM

The worm is a mature male. It is creamy white, delicate, filiform and wiry (Figs. 1, 2*A*). The anterior end is stiff and bluntly rounded. The posterior end is attenuate and exhibits a typical cork-screw coiling through at least three complete spirals. The total length of the filaria is 120 mm, the greatest diameter, 0.52 mm, and the diameter of the head, 0.41 mm. The integument is thick and is transversely striated, especially on the inner curvature of the tightly coiled posterior end. Second only

to the posterior spiralling, the most conspicuous feature of the worm is the relatively long ala on each side of the posterior extremity (Fig. 2*D*), with a length of 0.55 mm and a maximum breadth of 48 microns. Under low magnification of the uncompressed caudal end the 4 pairs of large preanal papillae were readily observed, while the 4 pairs of smaller postanal papillae were detected with considerable difficulty. For more detailed examination the posterior-most one mm was separated from the major portion of the worm with a sharp scalpel, mounted on a slide under a coverglass and studied under a magnification of 500 diameters (i.e., 4 mm objective and paired × 8 oculars).

FIG. 1. Photograph of entire male *Dirofilaria louisianensis* n. sp., natural size.

As observed either from the ventral or the lateral aspect (Fig. 2*D, E*) the caudal extremity showed clearly the 4 pairs of large, obovoidal, symmetrically arranged preanal papillae, approximately equal in size, each with a delicate central duct which apparently arises from a basal gland and opens externally through a pore. The 4 pairs of symmetrically arranged postanal papillae were considerably smaller. The two median pairs were structurally like the preanal papillae, but were only about half their size. The pair of papillae immediately posterior to the cloacal opening was even smaller, semi-stalked and its two members close together. Each papilla of the posteriormost pair resembled a delicate shoe-button on a moderately long, delicate stalk. All of these papillae were found to be smooth surfaced.

Cephalic papillae were difficult to detect but careful examination indicated that there were ten in all, consisting of four pairs of submedian

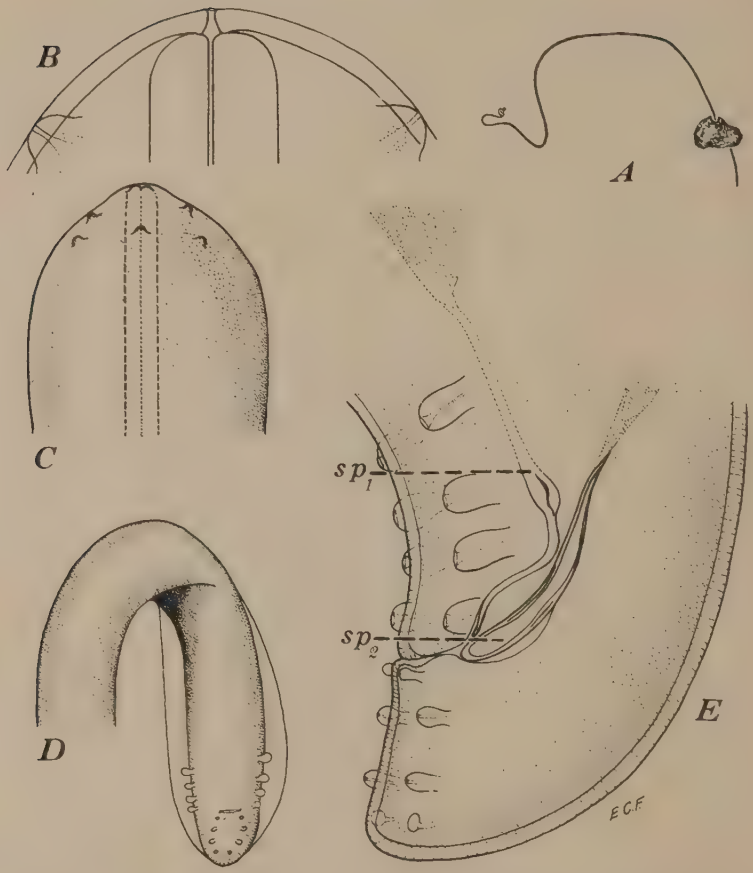


FIG. 2. *A*, camera lucida sketch of entire worm, with adherent portion of blood clot near anterior end, ×2/3; *B*, extreme anterior end of worm, lateral view, showing thickness of integument, two of the submedian papillae, proximal end of esophagus and minute buccal cavity, ×333; *C*, anterior end of worm, lateral view, showing contour of cephalic end, four submedian and one labial papillae, and portion of the esophagus, ×85; *D*, posterior end of worm, ventral view, uncompressed, showing caudal alae, cloacal cleft, 4 preanal papillae and 4 postanal papillae, ×85; *E*, posterior end of worm, nearly lateral view, slightly compressed, showing paired preanal and postanal papillae, long, narrow, left spicule (sp_1) and shorter, relatively broad, right spicule (sp_2), ×333.

papillae and one pair of lateral papillae (Fig. 2*C*). This conforms to the pattern presented by Chitwood and Chitwood (1938) for *Dirofilaria immitis*, which Desportes (1940), apparently unaware of the Chitwoods' data, identifies as a constant diagnostic character of the genus *Dirofilaria*.

The copulatory spicules (Fig. 2*E*, sp_1, sp_2) varied specifically from

TABLE 1.—*Comparative data on species of* Dirofilaria (*subgenus* Dirofilaria) (*Males only*)

	D. magalhãesi (from Brazilian boy)	*D. magalhãesi* (from Brazilian boy)	*D. louisianensis* n. sp. (from New Orleans negress)	*D. immitis* (from dogs)	*D. indica* (from Calcutta dog)	*D. pongoi* (from Borneo orang-outang)
Length, mm	83	83	120	120–200	130–170	107–125
Breadth, mm	0.28–0.40	0.407	0.52	0.6–0.9	0.7	0.33
Distance of cloaca from caudal tip, mm	0.11	0.11	0.33	0.075–0.12	0.110	0.095
Spicules, mm	Apparently single, 0.17 long	Shorter, 0.23; longer undoubtedly present(?)	Shorter, 0.150; longer, 0.196	Shorter, 0.2–0.226; longer, 0.3–0.355	Shorter, 0.165; longer, 0.385	Shorter, 0.18–0.195; longer, 0.306–0.312
Papillae: preanal	4 pairs, large, "villous," symmetrical	4 pairs, large, "villous," symmetrical	4 pairs, large, smooth, symmetrical	3–4 pairs, large, smooth, asymmetrical	4 pairs, large, "granular," bluntly digitate; one small papilla, median ventral; one pair, slender, adanal	4 pairs, large, club-shaped, having appearance of "pine cones"
" postanal ...	4 pairs, "villous," symmetrical; last 2 pairs smaller	4 pairs, "villous," symmetrical; last 2 pairs smaller	4 pairs, smooth, symmetrical; first and last pairs smaller	1 pair large, smooth, asymmetrical; 3–4 pairs, smooth, very small	9 pairs, some pedunculate, some sessile, varying in size but smaller than preanals	3–4 pairs, small digitate to conical; 1 pair very small, close together, immediately post-anal
Remarks	Based on study of worm by Blanchard (1896)	Based on French résumé sent by de Magalhães to von Linstow (1900)	Based on study of worm by present authors	Based on data of Fülleborn (1912), Vogel (1927), Faust (1937)	Based on study of worms by Chakravarty (1936)	Based on study of worms by Vogel and Vogelsang (1930)

those of described species of *Dirofilaria.* The right spicule of this worm (the shorter one), like that of other species of the subgenus, was seen to be fashioned as a moderately long, shallow, gracefully curved scoop, with a rounded distal end. On first examination its proximal (i.e. attachment) end appeared to be distinctly pointed, but more careful study revealed a nearly transparent, inverted triangular extension which constituted the actual internal shank of this spicule. It had a length measurement of approximately 150 microns, had ample skeletal support except at the proximal end, and was readily seen in the cleared specimen. The left spicule (the longer one) was more delicate, practically hyaline, and easily overlooked. It was not as sharply pointed at its distal end as that of *D. immitis* or *D. pongoi,* was curved on itself in its distal third to form a drawn-out figure S, had a distinct thickening at the level of flexure and a narrow shaft from this point to its proximal end, where it flattened out at its site of attachment. It had a length measurement of 196 microns.

The more important characters of this worm, together with comparable data from *D. magalhãesi, D. immitis, D. indica* and *D. pongoi* are summarized on Table 1.

There is no evidence associating this worm with any pathological procession in the host.

DISCUSSION

The discovery of a second isolated heartworm infection in man from an area far removed from that in which de Magalhães discovered the first human infection more than a half century ago indicates that man is susceptible to heartworm infection. However, since the human heart is almost invariably opened at autopsy, filarias, if present in the chambers of the heart or the great vessels leading into or from it, would probably not be overlooked, nor would they be diagnosed as artefacts. Thus, the absence of additional cases of human infection suggests that human heartworm infection is indeed rare.

If de Magalhães' worm and that from New Orleans were diagnosable as *Dirofilaria immitis,* an explanation of the two isolated human infections would be readily afforded, namely that this worm, which is so common a parasite of dogs, may, under appropriate circumstances, become a parasite of man. However, the present morphological criteria for species differentiation of filarioid worms require that the specimens obtained from the two human subjects be regarded as distinct from *D. immitis, D. indica* and *D. pongoi,* as well as from one another. Although it is apparent that parasites rarely reported from a particular host must have adequate reservoirs to maintain their life cycles, there is neither proof nor suggestive evidence indicating what these reservoirs are in the case of the two filarias recovered from the human heart.

The dilemma which results may be summarized as follows. From a standpoint of pure logic de Magalhães' worm and that from New Orleans should not be considered as distinct species, but until a broader species concept of filarioid worms is provided they must be designated as such.

Dirofilaria louisianensis n. sp.—with the characters of the genus *Dirofilaria,* subgenus *Dirofilaria.* Only a single male worm known, having a length of 120 mm, a greatest breadth of 0.52 mm and a distance from the cloacal cleft to the caudal extremity of 0.33 mm; with caudal alae 0.55 mm long and 48 microns wide; with 4 pairs of large, smooth, obovoidal, symmetrically arranged preanal papillae, approximately equal in size; with 4 pairs of relatively large, smooth, obovoidal, symmetrically arranged postanal papillae, of which the median two pairs are the larger and are similar in shape and structure to the preanal papillae, the first and fourth pairs are much smaller, the fourth pair having a shoe-button head surmounting a moderately long delicate stem; with a shorter (right) spicule, easily seen, measuring 0.150 mm in length, rounded at its distal end, with a nearly transparent proximal end; with the longer (left) spicule, delicate and nearly transparent so that it is not easily detected, measuring 0.196 mm in length, with an S-curved distal portion, a thickened flexure and a thin, flattened proximal end.

Habitat: New Orleans, La., U. S. A.

Host: Elderly negress, native and life-time resident of New Orleans.

Location of parasite in host: Inferior vena cava.

The specimen is deposited in the Faust Helminthological Collection.

SUMMARY

Dissection of the body of an aged negress, a native and life-time resident of New Orleans, La., has resulted in the recovery of a single adult male filaria from a blood clot in the inferior vena cava. The failure to find any females or microfilariae suggests that the male was the only worm that had developed. There was no evidence indicating that the heartworm infection had produced any pathology.

The measurements of this heartworm, together with the character of the preanal and postanal papillae and the structure of the copulatory spicules, indicate that on morphological grounds the worm is specifically different from the common heartworm of dogs (*Dirofilaria immitis*), from *D. indica,* from *D. pongoi* and from *D. magalhãesi,* the only previously reported heartworms of mammals. Based on these criteria this worm is designated as *Dirofilaria louisianensis* n. sp.

BIBLIOGRAPHY

BLANCHARD, R. 1895 Les Vers du Sang. In *Les Hématozoaires de l'Homme et des Animaux* (Paris), Pt. 2, 208 pp.

————1896 Parasites Animaux. In *Traité de Pathologie Générale* (Paris), pp. 782–783.

CHAKRAVARTY, G. K. 1936 *Dirofilaria indica* n. sp. from the heart of a dog. Z. Parasitenk. **9**: 57–60.

CHITWOOD, B. G. AND CHITWOOD, M. B. 1938 An Introduction to Nematology. Babylon, N. Y. Sec. I, Pt. 2, p. 61, Fig. 59A.

DE MAGALHÃES, P. S. 1887 Descripção de uma especie de filarias encontradas no coração humano, preçedida de uma contribuição para o estudo de filariose de Wucherer e do respectivo parasita adulto, a *Filaria Bancrofti*

(Cobbold) ou *Filaria sanguinis hominis* (Lewis). Rev. dos cursos praticos e theoricos da Faculd. de Med. do Rio de Janeiro, 3 ann., no. 3, pp. 193–214.

DESPORTES, C. 1940 Nouvelle description de l'extrémité céphalique de l'adulte de *Dirofilaria immitis* (Leidy, 1856). Ann. Parasitol. **17**: 405–414.

FAUST, E. C. 1937 Mammalian heart worms of the genus *Dirofilaria*. Festschrift Nocht, Hamburg. pp. 131–139.

VOGEL, H. AND VOGELSANG, S. G. 1930 Neue Filarien aus dem Orang-Utan und der Ratte. Z. Bakt., I. Abt., Orig. **117**: 480–485.

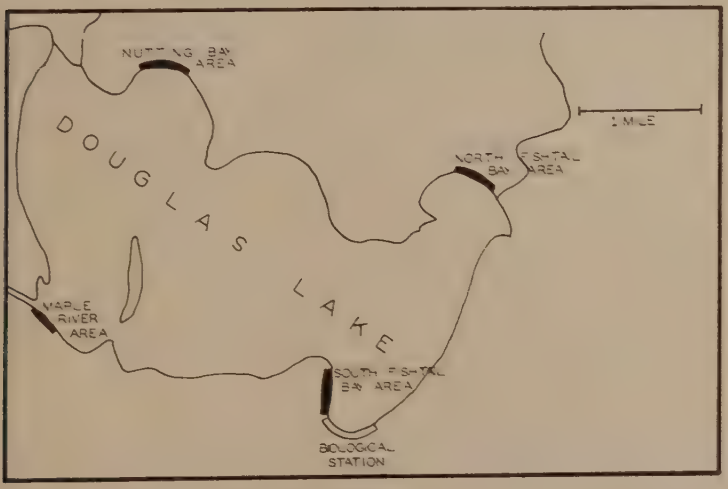
VON LINSTOW, O. 1900 Über die Arten der Blutfilarien des Menschen. Zool. Anz. **23**: 75–84.

LARVAL TREMATODE INFECTION IN JUVENILES AND ADULTS OF *PHYSA PARKERI* CURRIER[1]

W. W. Cort, Louis Olivier and D. B. McMullen

INTRODUCTION

Studies on the ecological relations of larval trematode infections in *Stagnicola emarginata angulata* (Sowerby) have been carried out during the last five summers at the University of Michigan Biological Station (see Cort, McMullen, and Brackett, 1937; Cort, McMullen, Olivier, and Brackett, 1940a). One of the most interesting parts of this work has been the comparison of infections in juvenile and adult snails. In the summers of 1938 and 1939 the ecological studies were extended to include the larval trematode infections of juveniles and adults of *Physa parkeri* Currier, a very large beach snail, and, in addition, its life cycle was worked out. Two preliminary publications have already been made on certain phases of this work (Cort, McMullen, and Olivier, 1938; Cort, McMullen, Olivier, and Brackett, 1940b). In this paper all the data obtained on the life cycle of *P. parkeri* and on its larval trematode infections are presented.



Map 1. Map of Douglas Lake showing the location of the four areas from which the collections of *P. parkeri* were made.

Received for publication, June 8, 1940.

[1] A contribution from the University of Michigan Biological Station and the Department of Helminthology, School of Hygiene and Public Health, The Johns Hopkins University, Baltimore, Maryland.

The collections of *P. parkeri* for this study were made from four areas on Douglas Lake (see Map 1). Of these, the North Fishtail Bay, Nutting Bay, and Maple River areas were also used in the studies of *S. emarginata angulata*. The collections of *P. parkeri* from the Maple River area, however, covered a much larger stretch of beach extending almost a half mile south and east of the source of Maple River. Here the beach is wide, the water is shallow, the soil contains much organic material, and there are many reed beds. In addition, collections were taken from the South Fishtail Bay area, which extends north about half a mile from the western limit of the Biological Station. The beach here varies considerably in width, but is narrow for most of the way. These four areas are among the best places for collecting *P. parkeri* in the whole region.

The methods used in this study were the same as those described by Cort et al (1940a).

LIFE CYCLE OF *Physa parkeri*

Physa parkeri is by far the largest species of this genus in the Douglas Lake region. Our measurements of adults (Table 1) show a range of shell length from 13–25 mm with the mode at about 20 mm.[2] These snails are very delicate and die quickly when brought into the laboratory unless handled very carefully and kept in running water. The shells, both of the juveniles and adults, are thin and easily broken in handling. This species rarely occurs in large numbers and appears to be easily affected by adverse conditions in its natural environment. *P. parkeri* has only been found on the shores of Douglas Lake. It is more restricted in its habitat than *S. emarginata angulata*, although it is sometimes found along stretches of beach where that variety is absent.

In the summer of 1939 we were able to make a few observations on the egg-laying habits of *P. parkeri*. From June 17 to 20 several egg masses were found and juveniles began to hatch from one of these on June 22. On June 20, July 4, and July 5 to 11, eggs were laid in the laboratory containers by snails brought in from three different areas. From four egg masses laid on June 20, juveniles hatched in 16 and 17 days, and during a warmer period (July 4 to 14) they hatched in from 8 to 10 days. The egg masses of *P. parkeri* are somewhat variable in size and shape and are usually larger than those of *S. emarginata angulata*. They are elongate, curved masses of jelly for the most part about 12 to 18 mm in length and are rather soft and easily broken. One unusually large mass was almost 50 mm long by about 12 mm wide. The differences in size depend on the number of eggs, which varied in 22 counts from 5 to 161. Most of the masses, however, contained from

[2] For the description of the methods used in making the measurements see Cort et al, 1940a, p. 35.

TABLE 1.—*Measurements (length in mm) of collections of juveniles and adults of* Physa parkeri *from Douglas Lake*

Length in mm	Maple River area						North Fishtail Bay area		South Fishtail Bay area			Nutting Bay area	
	Adults	Juveniles				Adults	Juveniles	Adults	Adults	Juveniles	Adults	Juveniles	
	7/11/38	7/11/38	7/27/38	8/13/38	9/3/38	6/15/39 6/22/39 7/14/39	8/28/38	6/17–20/39 7/10/39 7/20/39	7/25/38	8/22–23/38	6/14/39 6/23/39 7/3/39	7/23/38	8/26/38
3			2									1	
4			12									2	
5			20									3	
6			21							1		2	5
7			30	3						5		3	12
8			12	11						10		4	17
9			30	15			2			26			15
10			15	16	5		3			34			17
11		2	29	13	5		2			41			13
12		1	33	44	11		6			44			23
13		1	32	42	30		7			52	1		24
14		1	30	41	34	1	10			35	2		40
15			23	36	53	3	22			34	2		49
16	1		9	28	74	4	33			17	2		44
17	14		2	21	86	10	40	5	1	7	8		27
18	22			6	69	24	32	2	1	5	10		13
19	39			8	55	44	26	14	4	1	11		3
20	51			4	21	42	16	22	9	3	11		2
21	39				6	41	9	26	10	1	12		
22	17					23	1	30	13		1		
23	10					9	1	23	4		2		
24	4					2		11	5				
25	1							2	1				
Total	198*	5	300	318	449	203	210	135	48*	316	65	15	301

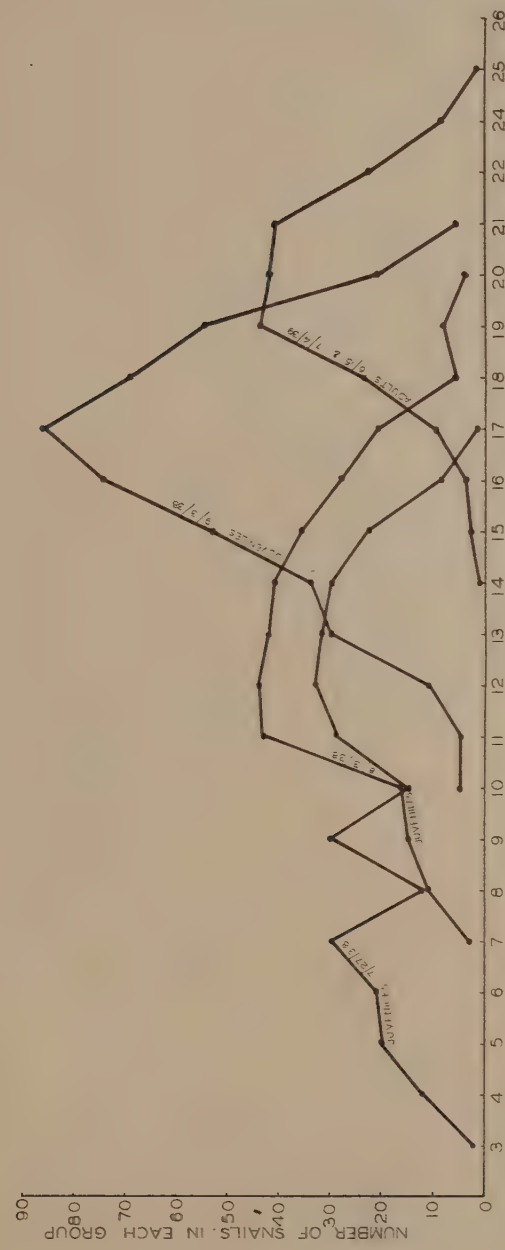
* These series include measurements of empty shells of snails that had recently died.

40 to 80 eggs. The newly hatched juveniles measured about 1 mm in length.

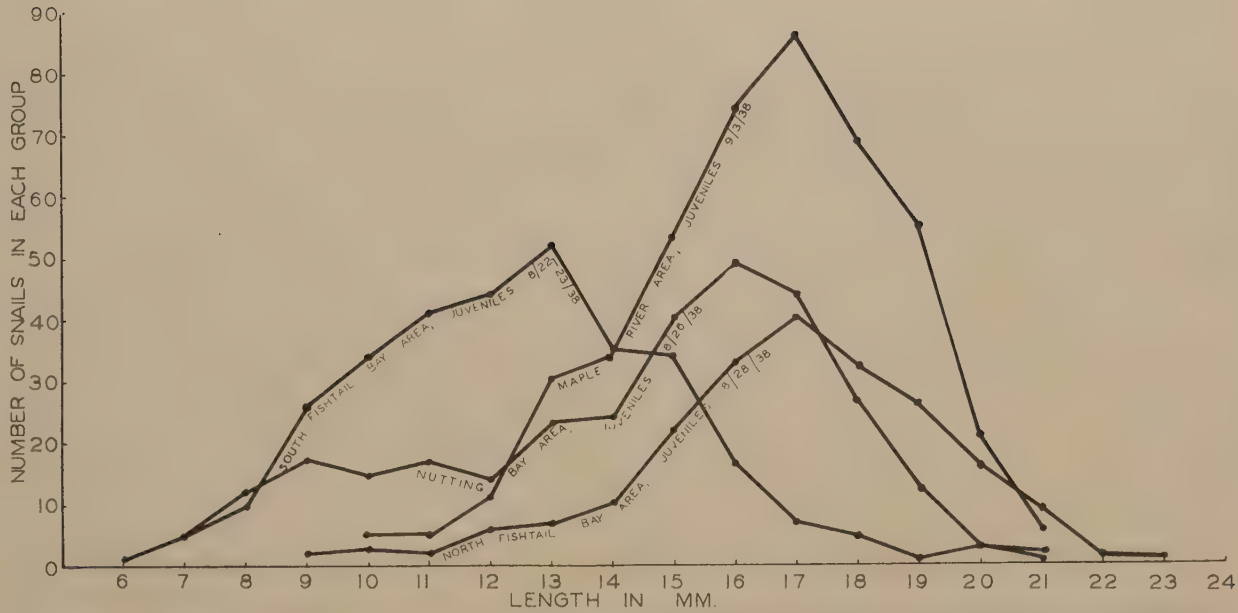
In the summer of 1938 we started to collect juveniles of *P. parkeri*. Unfortunately we have only a few records of early summer collections. On July 11 in making a collection of adults from the Maple River area we accidentally picked up five juveniles of this species from 11 to 14 mm in length. This suggests that on that particular beach some juveniles must have started to hatch in the first half of June or even earlier. Our best series of measurements of juveniles of *P. parkeri* was taken from specimens collected in the Maple River area on July 27, August 13, and September 3, 1938 (Table 1 and Graph 1). The 300 juveniles collected on July 27 had a surprising range of variation in length (3 to 17 mm) which overlapped the adult size. It seems probable that the smallest of these juveniles had hatched from eggs laid on the beach early in July. The considerable proportion of them that had reached a large size suggests that eggs of this species may be laid on the beaches somewhat earlier than those of *S. emarginata angulata*. By August 13 they had increased considerably in size, with more than half reaching a length greater than the minimum for adults (13 mm). None under 7 mm was present in this collection and there was a greater tendency for concentration of the length measurements around the mode. By September 3 all except a very few were in the adult size range. The minimum lengths in the three collections of juveniles shown in Graph 1 suggest that few eggs were being laid on this beach after the first week in July and that few if any snails were hatching after the middle of that month. We interpret this and the other evidence on the breeding season of *P. parkeri* as indicating that the eggs are laid in June and early July.

Late August collections of juveniles from the other three areas also all gave length measurements greatly overlapping those of adults. There was no evidence that any of these snails had reached sexual maturity, since no eggs were ever found. It can be suggested, therefore, that in the case of *P. parkeri* full size is reached in the fall and that growth is not resumed in the early spring of the second season. A comparison of the length measurements of the late summer collections of juveniles from the four different areas (Graph 2 and Table 1) suggests that in the South Fishtail Bay area growth was considerably slower than in the other three areas. Snails collected here on August 20 to 23 were smaller than those collected at approximately the same date from the three other areas.

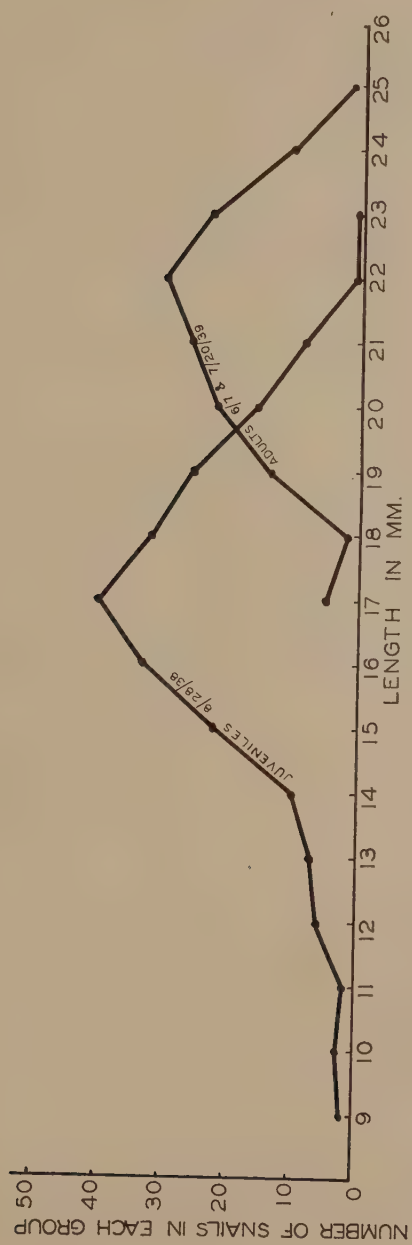
Growth between the time of the last fall collections and those made early the next June is shown in Graphs 1 and 3 for the snails from the Maple River and North Fishtail Bay areas and the same thing for the South Fishtail Bay area can be seen in Table 1. On the basis of these



GRAPH 1. Measurements of shell length of juveniles of *P. parkeri* from collections from the Maple River area on July 27, August 13, and September 3, 1938, compared with those of adults of the same generation collected in the early summer of 1939 (see Table 1).



GRAPH 2. A comparison of the length measurements of collections of juveniles of *P. parkeri* from the four areas made during the last half of the summer of 1938 (see Table 1).



GRAPH 3. Length measurements of juveniles of *P. parkeri* from a late summer collection in 1938 from the North Fishtail Bay area compared with those of adults from the same area collected early in the summer of 1939 (see Table 1).

data it seems reasonable to conclude that the full growth could easily have been attained in the fall before cold weather set in. It is also evident that there must have been a slowing up in the growth of the larger juveniles in all three areas, since the range of variation in the adults is less than in the juveniles of the previous summer. Here it can be suggested, as was done earlier for *S. emarginata angulata* (Cort et al, 1940a), that adult size may have been fixed by the beginning of cold weather, that the largest juveniles became the largest adults, and that the smallest juveniles remained small as adults. This suggests also the possibility that an important cause of variation in the adults, as in the juveniles, is the time of egg laying. According to this view, in general, those snails that hatched early in the summer became the largest adults and those from eggs laid late in the breeding season became the smallest adults.

The records of collections in Table 1 show that the adults of *P. parkeri* were scarcer on the beaches in the summer of 1939 than in 1938. It seems probable that a combination of factors led to this reduction in the numbers. In the Maple River area there was evidence that the juvenile snails were being killed in large numbers by exceptionally heavy larval trematode infections. The collections of the summer of 1938 were probably also a factor, especially in the South Fishtail Bay area, where we deliberately tried to collect out the snails because of danger of schistosome dermatitis. Other factors, probably adverse environmental changes, seem to have been operating in this reduction, especially in the Nutting Bay area, where the incidence of trematode infections was low in 1938 and where little collecting was done that summer.

Our collections on the beaches of Douglas Lake over a period of years had given the general impression that adults of *P. parkeri* are scarce after the middle of the summer and the collections of the summers of 1938 and 1939 confirmed this impression. Even in the earliest collections large numbers of empty shells were found, which showed from their color and condition that they belonged to adult snails that had recently died. Also, on every collecting trip we found shells which still retained the dead bodies of snails. In addition, it became increasingly hard to find adult snails in collections made after the middle of July. It is evident, therefore, that soon after the end of the breeding season, the adults of *P. parkeri* die and are almost entirely replaced by the new generation of juveniles by August.

The life span for *Physa parkeri,* therefore, is about 13 to 14 months, and extends over only one winter. Egg laying begins sometime in June and few if any eggs are laid after the middle of July. The juveniles grow very rapidly, the majority overlapping the adult size by the end of August, but do not attain sexual maturity until the next spring after pass-

ing the winter under the ice. After the breeding season the adults soon die, are scarce by the middle of July, and appear to be all gone soon after the first of August. As compared with *Stagnicola emarginata angulata,* the breeding season of *P. parkeri* appears to be somewhat earlier, the juveniles attain a relatively larger size by the end of the summer, and the adults die sooner in the summer.

LARVAL TREMATODE INFECTIONS IN JUVENILES AND ADULTS OF *Physa parkeri*

In the four areas on Douglas Lake *P. parkeri* harbors six species of larval trematodes (see Table 2), viz., a schistosome (*Cercaria physellae* Talbot, 1936), an echinostome (the cercaria of *Echinoparyphium recurvatum* (Linstow, 1873)), a strigeid (*C. physae* Cort and Brooks, 1928), a notocotylid (the cercaria of *Notocotylus urbanensis* (Cort, 1914)), and two plagiorchiids (the cercaria of *Cercorchis medius* (Stunkard, 1916) and one belonging to the Reniferinae).

C. physellae is one of the dermatitis-producing schistosome cercariae of the region. It is found also in the small beach *Physa, P. magnalacustris,* and in the Douglas Lake region seems to be pretty well limited to these lake shore physae. It has been reported by Brackett (1940a) from other species of this genus in Wisconsin and is found there in a wide variety of habitats. In the Douglas Lake region it seems to be of little significance in the production of the "swimmer's itch" on the bathing beaches (Cort et al, 1940b). The adult of *C. physellae* is not known but the recent epidemiological studies of Brackett (1940b) in Wisconsin indicate that its definitive host is probably a duck.

The cercaria of *Echinoparyphium recurvatum* had a high incidence in our collections. Its adult has a wide range of hosts, both birds and mammals. It is common in the muskrats in the Douglas Lake region and its high incidence in the Maple River area (Table 2) was attributed to the presence on this beach of large numbers of these hosts. The cercaria of this species is also present in other species of *Physa* including those, such as *P. gyrina,* that live in ponds and swamps. It is also found in low incidence in *Stagnicola emarginata angulata.*

The strigeid, *C. physae,* is very similar to the cercaria of the heron strigeid, *Posthodiplostomum minimum* (Hunter, 1936 and 1937; Ferguson, 1936 and 1937). This cercaria is very widespread in the region and is found commonly in various species of *Physa,* including those from small ponds and swamps. It seems probable that its definitive host may be the great blue heron which is so common in the Douglas Lake region.

The notocotylid of our collections was shown experimentally by Herber (1939) to be the cercaria of *Notocotylus urbanensis.* The cercaria of this species is also widely distributed in the various species of *Physa* and is present in low incidence in *S. emarginata angulata.*

TABLE 2. *Larval trematode infections in* Physa parkeri *collected from four areas on Douglas Lake during the summers of 1938 and 1939*

	Maple River area				South Fishtail Bay area			North Fishtail Bay area			Nutting Bay area
	Adults	Juveniles		Adults	Adults	Juveniles	Adults	Adults	Juveniles	Adults	Juveniles
	7/11/38 7/27/38	7/27/38	9/3/38	6/15/39 6/21/39 7/ 4/39	6/18/38 7/ 7/38 7/21/38 7/25/38	8/22– 23/38	6/14/39 6/23/39 7/ 3/39	6/21/38	8/28/38	6/17–20/39 7/10/39 7/20/39	8/26/38
Total collection	150	298	440	203	355	322	66	60	209	137	300
Negatives	25 (17)*	110 (38)	291 (66)	120 (59)	293 (83)	304 (94)	58 (88)	35 (59)	157 (75)	72 (54)	260 (87)
Immature (unidentified positives)	0	3	1	0	3	0	0	0	0	3	0
Multiple infections	4	2	1	0	0	0	0	1	1	0	0
Cercaria physellae—Total	24 (16)	1 (1)	16 (4)	31 (17)	34 (10)	1	5 (8)	18 (30)	5 (2)	35 (26)	0
Old	17	1		2	18			8			
Mature	7	2	15	27	13	1	5	10	4	25	
Young mature		1	1		3				1	4	
Immature				5						6	
Cer. *E. recurvatum*—Total	82 (55)	170 (57)	84 (19)	31 (17)	11 (3)	1	0	4 (7)	14 (7)	11 (8)	3 (1)
Mature	59	100	83	32	11	1		1	6	9	2
Immature	23	70	1	2					8	2	1
C. physae—Total	4 (3)	12 (4)	37 (8)	9 (4)	2 (1)	15 (5)	0	0	23 (11)	7 (5)	10 (3)
Old				1		1					
Mature	1	10	34	4	2	7			17	6	7
Young mature						6			3	1	2
Immature	3	2	3	4		1			3		1
Cer. *N. urbanensis*—Total	17 (11)	1	8 (2)	3 (2)	12 (3)	1	3 (5)	4 (7)	3 (1)	7 (5)	27 (9)
Mature	17	1	7	3	12	1	3	4	3	5	14
Young mature											7
Immature			1							2	6
Cer. *C. medius*—Total	2 (1)	0	4 (1)	0	0	0	0	0	0	0	0
Mature	2		4								
Cer. Reniferinae—Total	0	0	0	3 (2)	0	0	0	0	8 (4)	2 (2)	0
Mature				2					8	2	
Immature				1							

* Percentage incidence in collection.

The cercaria of *Cercorchis medius*, a turtle telorchiid (McMullen, 1934), was described from *Physa integra* and occurred in *P. parkeri* from only one of our areas.

The reniferid cercaria found in two of the collections is probably one of the two species for which the life cycles were described by Talbot (1933). As noted by this author it is almost impossible to make accurate identifications of these species from the cercaria alone. These cercariae are widely distributed and occur in several species of *Physa*. Their definitive hosts are garter snakes.

The incidence of infection of these six species of cercariae in *P. parkeri* is such that multiple infections would not be expected to be common except in the Maple River area (Cort et al, 1937). In fact in the whole series of examinations listed in Table 2 only nine double infections were found. *C. physellae* was found in three cases with the cercaria of *N. urbanensis*, once with the cercaria of *E. recurvatum*, once with *C. physae*, and once with an infection too young to be identified with certainty (probably *C. physae* since it had elongate daughter sporocysts). Also, the cercaria of *E. recurvatum* was found twice with that of *N. urbanensis* and once with the reniferid cercaria. An analysis of the chances of combinations of these species of cercariae occurring in double infections suggests that certain combinations must have been prevented by an immunity or antagonism. For example, there were no double infections of the cercaria of *E. recurvatum* and *C. physae*, although according to chance they should have occurred between 15 and 20 times in the collections from the Maple River area. Also *C. physellae* was found in only one double infection with the cercaria of *E. recurvatum* in the collections from this same area when according to chance this combination should have been found over 20 times.

The evidence indicates that juveniles of *P. parkeri* are infected with larval trematodes at all stages of development, that infections are carried over the winter in large numbers, and that adults also acquire infection after they pass the winter under the ice. The data on the infection of very young juveniles, which came from the collection of July 27 from the Maple River area are of special interest. From this collection the juveniles were individually measured at the same time they were examined for larval trematodes. Immature infections of the cercaria of *E. recurvatum* were found in four snails 3 and 4 mm in length and mature infections in nine individuals only 5 and 6 mm long. Also a mature infection of *C. physae* was present in a snail 6 mm in length, an old infection of *C. physellae* in one 9 mm, and a mature infection of *Neocotylus urbanensis* in one 10 mm. Other collections gave a small amount of similar evidence (Table 2). These findings, especially the presence in rather small snails of so many mature infections, which must have taken

a minimum of five to six weeks to develop, suggest that the miracidia penetrate the juveniles of this snail soon after they hatch from the eggs.

It is interesting to compare the infections with the cercaria of *E. recurvatum* in juveniles of different sizes collected from the Maple River area on June 27, 1938 (Table 3). In this analysis it appears that the

TABLE 3.—*Infection with the cercaria of* E. recurvatum *in different sized juveniles of* P. parkeri *collected in the Maple River area on July 27, 1938*

	Length in mm							
	3 and 4	5	6	7	8	9	10	11 to 17
Number in group	14	20	21	30	12	30	15	156
Negatives	9	11	11	18	6	14	5	36
Cer. *E. recurvatum:*								
Immature	4	4	4	6	4	5	3	40
Mature	0	5	4	4	1	10	5	71
Total	4	9	8	10	5	15	8	111

smaller snails tend to have a larger proportion of negative, and the proportion of immature to mature infections became progressively less in the larger snails. It seems evident, therefore, that the larger juveniles were older and consequently had a longer period of exposure to infection.

Evidence that infections of the juveniles are carried over the winter in the snails comes from the June collections of adults made in 1938 and 1939. In 1938 the ice broke up on Douglas Lake on April 12 and in 1939 on April 25 to 28. As noted earlier (Cort et al, 1940a) the temperatures of April and early May in this region are almost too low for development of larval trematodes and development would be slow until late in June. It seems almost certain, therefore, that at least the old and mature infections with larval trematodes that were found in adult snails in June (Table 2) had entered the previous season and had been carried over the winter.

There is, in addition, evidence of susceptibility of adults of *P. parkeri* to infection with larval trematodes. For example, in a collection of 114 adults from the Maple River area on July 11, 1938, there were 21 immature infections with the cercaria of *E. recurvatum.* Also, it can be noted from Table 2 that in the collections of adults from all the areas there was a considerable number of immature infections representing all but one of the species of larval trematodes. In addition, there were six infections too immature for identification. It, therefore, becomes evident that some adults of *P. parkeri* were being infected during their second season.

Some evidence concerning the time that infections with the different species of larval trematodes were acquired and their course in the snail hosts is found in Table 2. The low incidence made it impossible to get any evidence on these points for the two plagiorchiid species. Of the larval stages of *N. urbanensis* also little can be said, except that few im-

mature infections were found and the adult snails in general had higher incidences than the juveniles.

C. physellae. As brought out in an earlier paper (Cort et al, 1940a) the development period for schistosomes from the time the fully embryonated eggs are dropped into the water until the first cercariae escape from the infected snails would be, in the Douglas Lake region, about 5 to 6 weeks in the warmest part of the summer. A total of 76 infections of *C. physellae* was found in adult snails collected in June and July, 1938, from three areas. Of these, 43 infections were listed as old, 30 as mature, and 3 as young mature. In June the proportion of old infections was about 50 and in July about 60 per cent. It seems probable that all these old and mature infections had entered the snails the previous season, probably both late in the summer and early in the fall. The incidence of infection with *C. physellae* in the juveniles of *P. parkeri* was very low as compared with that of the adults. In the Maple River area there were only 4 infections in almost 300 juveniles collected July 27, 1 old, 2 mature, and 1 young mature. Even postulating the minimum possible time of development it is evident that these infections must have entered the snails when they were very small. Some infection of snails later this same summer is indicated by the presence of 15 mature and 1 young mature infection in the juveniles collected from this area on September 3. The adult snails collected in the early summer of 1939 in this area showed a greatly increased incidence of infection with *C. physellae* as compared with the juveniles of the previous summer from which they developed. When we consider that the season in 1939 was unusually late it seems almost certain that all these infections, except the five immature ones, had entered the juveniles the previous season. In the late August (1938) collections of juveniles from the South Fishtail Bay area only one infection of *C. physellae* was present and in that from the North Fishtail Bay area there were 5 (4 mature and 1 immature). Adults collected from the North Fishtail Bay area for the next summer showed a great increase in the incidence of *C. physellae.* While most of these infections probably came in the previous fall, the 6 immature infections which were found in the June 17–20, 1939, collection may have come in the same season.

Although the adult stage of *C. physellae* is not known, it seems almost certain, as indicated above, that it lives in a species of duck. These hosts must have been giving off the eggs of this schistosome on the beaches of Douglas Lake chiefly in the late summer and early fall to produce the large number of infections that were carried over the winter. Some slight infection of snails also occurred in the spring and first half of the summer as indicated by the infections found in juveniles. These relations and the characteristics of the life cycle of *P. parkeri* indicate

that the only time of the year that the cercariae of this schistosome would be escaping from its snail host in sufficient numbers to produce outbreaks of dermatitis in man would usually be in May, June, and early July before the swimming season got well under way. Even by the end of June the number of adult snails is considerably reduced by natural death and in a season such as 1938 a considerable proportion of the infections of *C. physellae* would have exhausted their supply of cercariae. The slight infection found in the juveniles would not be sufficient to produce enough cercariae to make them a factor in the dermatitis. Also the snails of this species are seldom numerous on the beaches. The fact is that only in one area where several people contracted the dermatitis early in the summer have we obtained any evidence of "swimmer's itch" produced by *C. physellae.* Therefore, this schistosome cercaria, as was suggested in an earlier paper (Cort et al, 1940b), can be almost entirely disregarded in relation to the problem of "swimmer's itch" in the Douglas Lake region.

C. physae. The data available in the literature (see van Haitsma, 1930, and 1931a and b) indicate that in strigeids the time of development from the dropping of eggs into the water until the first cercariae escape from the infected snails is about 8–10 weeks under favorable temperatures, the development in the snail requiring about 6 to 7 weeks. If we accept this time for the development of *C. physae* the data from our examinations (Table 2) suggest that infection of snails with this species takes place at various times during the "open" season. Immature infections in adult snails of the June and July collections suggest that they become infected in the late spring or early summer. Also, the finding of mature infections in juveniles in the collection of July 27, 1938, from the Maple River area indicates that the eggs of this species were dropped into the water in the spring and that the snails were infected in June. The presence of mature, young mature, and immature infections in juveniles collected from all four of our areas from August 23 to September 3 further shows that snails were infected during June, July, and perhaps early August. Mature infections in the adult snails were almost certainly derived from infections that were carried over the winter. It seems evident, therefore, that the definitive host of *C. physae* must visit the beaches and deposit the eggs in the water at various times in the spring, summer, and fall. The great blue heron, which was suggested above as a possible definitive host for *C. physae,* frequents the beaches throughout the summer, so that an adult strigeid in this host might well produce the kind of snail infection that was found for *C. physae.*

Another very interesting point about *C. physae* is that the incidence of infection is much higher in juveniles than in adults. In the three areas where we have late summer records for juveniles followed by examina-

tions of the adults next season, the incidence of *C. physae* in the latter has been greatly reduced (see Table 2). In the late summer collections of juveniles most of the infections with *C. physae* were already mature. What seems to us the most reasonable explanation for the lower incidence in adult snails is that infections with this cercariae that mature in the juvenile snails during their first summer become old during the fall, and disappear sometime during the winter, very probably by the death of the infected snails. This view is supported by the fact that only one old infection of *C. physae* was found in an adult snail. Infections of strigeids when they become old almost completely destroy the digestive glands of the infected snails and would naturally be injurious to growing juveniles. Evidence was also presented in an earlier publication (Cort et al, 1939) that infections of the gull strigeid, *Diplostomum flexicaudum,* cause the death of numbers of juvenile *S. emarginata angulata.*

Cercaria of Echinoparyphium recurvatum. The data on this species (Table 2) show infection of high incidence in both juveniles and adults. It is improbable that the juveniles of *P. parkeri* 5 to 6 mm in length that harbored mature infections of this larval trematode on July 27, 1938, on the beach in the Maple River area were more than a month old. It can be suggested, therefore, that the development of this species in the snail host under unusually favorable conditions can be completed in about a month.

As stated above we concluded that the large amount of infection in the snails of the Maple River area came from the muskrats. In the first collection of *P. parkeri* from this beach (July 11, 1938) sixty of the 114 adults were infected with this echinostome species (39 mature and 21 immature). On July 27, 36 adults were taken from the same beach. Twenty-two of these snails were infected with the echinostome but only two of the infections were immature. On this same date 298 juveniles, 3 to 17 mm in length, were also collected, of which 170 were infected with the cercaria of *E. recurvatum.* Seventy of these infections were immature. Thus large numbers of both juveniles and adults of *P. parkeri* were becoming infected with this echinostome early that season, although it is possible that the mature infections of the adults of the July 11 collection had in part at least been carried over the winter.

We noted in the examinations of the juveniles of the July 27 collection that the digestive glands of the snails were very severely damaged especially by the mature infections in which almost all the tissue was replaced by numbers of large rediae. The juveniles were very numerous on this beach on July 27 and our collection of about 300 snails reduced the numbers only slightly especially since we covered only a part of the beach. When we made our collection of September 3 the number of juveniles was much less than on the earlier date although they were

still fairly numerous and we were able to collect 440 in about three hours. The examination of this collection showed a very great reduction of the incidence of the cercaria of *E. recurvatum* from 57 per cent on July 27 to 19 per cent on September 3. This very striking reduction in incidence certainly suggests that the infections with this larval trematode were killing large numbers of these snails and were probably a significant factor in reducing their numbers. Also, the fact that 83 of the 84 infections of the cercaria of *E. recurvatum* found in the juveniles on September 3 were mature indicates that the infection of the snails was practically limited to the first part of the summer although the reason for this is not clear. When collections of adults of *P. parkeri* were made early the following summer, it was found that they were much scarcer than they had been the previous summer. As suggested earlier in this paper, this reduction in numbers was probably due to a large extent to the killing of great numbers of the juveniles by the echinostome infections. It seems probable also that most of the echinostome infections in these snails collected in 1939 had lived over the winter since only 2 immature infections were present.

While many workers have called attention to the injury to the snail hosts produced by larval trematode infections only a few reports present definite evidence indicating that they are an important lethal factor under natural conditions. Johnson (1920, pp. 363–364) suggested that infections with the cercaria of *Echinostoma revolutum* were an important cause of the death of the snails in a heavily infected pond where he made his collections. Wesenberg-Lund (1934, pp. 183–184) has also emphasized the importance of larval trematode infections in the reduction of snail populations. Our own data indicate particularly the death of juvenile snails brought about by infections of larval trematodes introduced so early that they are able to run their course and grow old before the maturity of the snail. The digestive glands of the snails are almost completely destroyed by old infections, especially of echinostomes, schistosomes, and strigeids, where the rediae and daughter sporocysts invade all the tissue. We are of the opinion that snails are unable to recover from such infections. They probably shorten the lives of adult snails, but seem to be especially important in their effects on snail populations by killing large numbers of juveniles before the breeding season.

SUMMARY

Investigations were made during the summers of 1938 and 1939 on the relations of larval trematode infections to the large beach *Physa, P. parkeri*, in four areas on Douglas Lake, Michigan. The life span of this snail species appears to be from about 13 to 14 months. The eggs are laid by the adults in the late spring and early summer, probably from

late May to early July. The period of embryonic development varies from 8 to 17 days according to the temperature and the newly hatched juveniles are about 1 mm in length. The juveniles grow rapidly on the beaches during the summer and by late August greatly overlap the adult size. They probably reach adult size in the fall. After passing the winter under the ice they are all sexually mature early the next summer. Soon after their breeding season, even before the end of June, they begin to die off rapidly and almost all are gone before the end of July. Therefore, collections of *P. parkeri* made at the end of the summer contain only large juveniles, while those of early June contain only sexually mature adults.

In the collections of both juveniles and adults of *P. parkeri* from the four study areas on Douglas Lake six species of larval trematodes were found, viz., a dermatitis-producing schistosome cercaria (*Cercaria physellae* Talbot, 1936), an echinostome (the cercaria of *Echinoparyphium recurvatum* (von Linstow, 1873)), a strigeid (*C. physae* Cort and Brooks, 1928), a notocotylid (the cercaria of *Notocotylus urbanensis* (Cort, 1914)), and two plagiorchiids (the cercaria of *Cercorchis medius* (Stunkard, 1914) and one belonging to the Reniferinae). Only 9 double infections were found involving 5 combinations of these six species of cercariae. It is suggested that there is some antagonism or immunity relation that prevents combinations in double infections of the echinostome with the schistosome and strigeid since each of these combinations would have otherwise occurred much more often.

The finding of adult infections in rather small juveniles suggested that they were infected when very young, probably soon after hatching. A large proportion of the infections found in the adult snails appears to have been carried over the winter, although the evidence is clear that some infections enter the adults early the second summer. In the case of the schistosome, *C. physellae,* it seems probable that most of the infections enter the juveniles in the late summer and early fall and are carried over the winter, because the incidence of the infections is much lower in the juveniles than in the adults.

Infections of the strigeid, *C. physae,* appear to enter the snails all through the open season on the lake, since immature infections were found in almost all collections, both of juveniles and adults. The great reduction of incidence of *C. physae* in the adult snails as compared with the juveniles suggests that infections that come to maturity in the juveniles kill them in considerable numbers.

The very high incidence of the cercaria of *E. recurvatum* in one area was attributed to the abundance of muskrats. This cercaria infected very young juveniles and came to maturity in numbers in those that were only partly grown. These infections apparently killed large num-

bers of the juvenile snails in this area. This and other species of larval trematodes, especially strigeids and schistosomes, as they grow older almost completely destroy the digestive glands of the infected snails. It seems probable, therefore, that snails never recover from such infections, and that wherever they mature in numbers in juveniles, they may be an important lethal factor which may affect adversely whole snail populations.

Bibliography

Brackett, Sterling 1940a Studies on schistosome dermatitis. V. Prevalence in Wisconsin. Am. J. Hyg. **31** (Sec. D) : 49–63.

———1940b Studies on schistosome dermatitis. VIII. Notes on the biology of the snail hosts of schistosome cercariae in Wisconsin and epidemiological evidence of the life cycles of some avian schistosomes. Am. J. Hyg. **32** (Sec. D) : 85–104.

Cort, W. W., D. B. McMullen and Sterling Brackett 1937 Ecological studies on the cercariae in *Stagnicola emarginata angulata* (Sowerby) in the Douglas Lake region, Michigan. J. Parasitol. **23** : 504–532.

Cort, W. W., D. B. McMullen and Louis Olivier 1938 Larval trematode infections in juveniles of *Physa parkeri* Currier. J. Parasitol. **24** Suppl : 25–26.

Cort, W. W., D. B. McMullen, Louis Olivier and Sterling Brackett 1939 The larval stages of the gull strigeid, *Diplostomum flexicaudum,* in juveniles and adults of *Stagnicola emarginata angulata.* J. Parasitol. **25** Suppl : 8.

———1940a Studies on schistosome dermatitis. VII. Seasonal incidence of *Cercaria stagnicolae* Talbot, 1936, in relation to the life cycle of its snail host, *Stagnicola emarginata angulata* (Sowerby). Am. J. Hyg. **32** (Sec. D) : 33–69.

———1940b Recent studies on schistosome dermatitis in the United States. Rev. Med. Trop. y Parasitol., Bact., Clin. y Lab. **6** : 55–64.

Ferguson, M. S. 1936 Experimental studies on *Neascus vancleavei.* J. Parasitol. **22** : 544–545.

———1937 Experimental studies on *Posthodiplostomum minimum* (MacCallum, 1921), a trematode from herons. Thesis Abstract, Dept. Zool., Univ. Illinois, pp. 1–7.

Herber, E. C. 1939 Life history studies on monostomes of the genus *Notocotylus* (Trematoda). J. Parasitol. **25** Suppl : 18–19.

Hunter, G. W. III. 1936 Penetration of the common sunfish by the holostome cercaria, *C. multicellulata.* J. Parasitol. **22** : 542.

———1937 Parasitism of fishes in the lower Hudson area. Suppl. 26th Ann. Rept. N. Y. State Cons. Dept. Biol. Surv. No. XI. Lower Hudson Watershed, 1936, pp. 264–273.

Johnson, J. C. 1920 The life cycle of *Echinostoma revolutum* (Froelich). Univ. Calif. Publications Zool. **19** : 335–388.

McMullen, D. B. 1934 The life cycle of the turtle trematode, *Cercorchis medius.* J. Parasitol. **20** : 248–250.

Talbot, S. B. 1933 Life history studies on trematodes of the subfamily Reniferinae. Parasitology **25** : 518–545.

Van Haitsma, J. P. 1930 Studies on the trematode family Strigeidae (Holostomidae). XXI. Life cycle and description of the cercaria of *Cotylurus michiganensis* (La Rue). J. Parasitol. **16** : 224–230.

———1931a Studies on the trematode family Strigeidae (Holostomidae). No. XXII. *Cotylurus flabelliformis* (Faust) and its life history. Papers Michigan Acad. Sc., Arts and Letters **13** : 447–482.

———1931b Studies on the trematode family Strigeidae (Holostomidae). No. XXIII. *Diplostomum flexicaudum* (Cort and Brooks) and stages in its life history. Papers Michigan Acad. Sc., Arts and Letters **13**: 483–516.

WESENBERG-LUND, C. 1934 Contributions to the development of the Trematoda Digenea. Part II. The biology of freshwater cercariae in Danish freshwater. Danske Vidensk. Selsk. Skr. Naturv. og Math. Afd. (Mem. Acad. Roy. Sc. et Lett. Danemark, Copenhagen, Sec. Sc.) **5**: 1–223.

STUDIES ON PHENOTHIAZINE. IX. THE BILIARY EXCRETION AND ANTHELMINTIC ACTION OF THIONOL

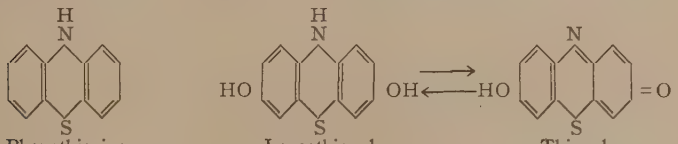
FLOYD DEEDS AND JOHN O. THOMAS

Bureau of Agricultural Chemistry and Engineering, United States Department of Agriculture, at the Department of Pharmacology, Stanford University School of Medicine, San Francisco, California*

Phenothiazine, described by Smith, Munger, and Siegler (1) in 1935, is one of the most effective, and certainly the most versatile, of the organic compounds developed by the U. S. Department of Agriculture. In addition to being a promising insecticide phenothiazine has been shown to be useful as a urinary antiseptic (2), and is now receiving considerable attention as an anthelmintic (3–7) for removal of parasites from farm animals, notably sheep.

In a previous paper (8), proof of the oxidation of phenothiazine to thionol after oral administration of phenothiazine to rabbits, rats, and man was given, and the excretion of thionol, leucothionol, and phenothiazine was discussed. At that time interest was limited largely to the urinary excretion of phenothiazine and its oxidation products because of the bactericidal properties (9) conferred upon the urine.

Recent observations have demonstrated the presence of phenothiazine, leucothionol, and traces of thionol in the feces, the oxidation product existing predominantly in the leuco form because of the reducing properties of the intestinal contents. In view of the bactericidal properties of thionol, the presence of phenothiazine, thionol, and leucothionol in the intestine has acquired a new interest following the reports of the anthelmintic action (3–7) of orally administered phenothiazine. The structural relationship of these compounds is shown in the following formulae:



Phenothiazine Leucothionol Thionol

If the conversion of phenothiazine to thionol and leucothionol, at least in part, takes place in the digestive tract, the presence of these three substances in the feces could be explained on the basis of incomplete absorption. If absorption from the digestive tract is complete, or if oxidation of phenothiazine takes place only after absorption, the presence of these

Received for publication, July 1, 1940.
* Food Research Division Contribution No. 500.

compounds in the intestinal tract indicates an excretion into the intestine through the biliary tract.

EXCRETION IN RABBITS

To obtain information on the possibility of biliary excretion in this species two rabbits were used. One rabbit, weighing 3.7 kilograms, was given 3 grams of phenothiazine by stomach tube, and the other, weighing 3.5 kilograms, was given 3 grams of thionol. The rabbits were killed about six hours later and the bile removed from the gall bladders by means of a hypodermic syringe. Each bile sample was treated as follows: After diluting it with about 100 cc of distilled water and warming, concentrated hydrochloric acid was added until no further precipitation occurred. The sample was then filtered and extracted with ether in a separatory funnel. After evaporation of the ether the residue was dissolved in a small volume of 95 per cent alcohol, which yielded a red solution. The alcoholic solution was then diluted with water. Upon the addition of water the sample obtained from the rabbit given phenothiazine yielded a fine silky-appearing precipitate, indicative of unoxidized phenothiazine, which was removed by centrifuging. The supernatant dilute alcohol was now poured off. This reddish supernatant fluid gave an intensification of the red color upon the addition of either hydrogen peroxide or sodium hydroxide, thus indicating that both thionol and leucothionol were present in the bile. The sample of bile from the rabbit receiving thionol gave the same results, except that there was, of course, no silky precipitate indicative of phenothiazine. Examination of control bile gave negative results.

The positive results obtained demonstrate that the gastric administration of phenothiazine to these rabbits resulted in the biliary excretion of phenothiazine, thionol, and leucothionol, and that the gastric administration of thionol gave rise to the presence of thionol and leucothionol in the bile.

EXCRETION IN DOGS

Biliary excretion of thionol in this species was demonstrated with three animals. Three dogs weighing 14.8, 12.5, and 10.8 kilograms were given 1.75, 1.75, and 1.5 grams of phenothiazine respectively in gelatin capsules. Six hours later when these dogs were operated upon in the laboratory of experimental surgery, bile samples were obtained with sterile hypodermic syringes. The bile samples were examined as outlined for the experiment on rabbits. Each of the three samples of bile gave positive evidence for the presence of thionol, leucothionol and traces of phenothiazine.

EXCRETION IN MAN

While treating patients having urinary tract infections with orally

administered phenothiazine, Dr. A. B. Stockton of the Department of Medicine encountered a patient with a biliary fistula. This fortuitous circumstance permitted the collection of a sample of bile before and after the administration of phenothiazine. The human bile thus obtained was examined as outlined for rabbits. The control sample of bile, obtained prior to administration of phenothiazine, gave negative tests, but the sample following use of the drug gave positive evidence for the presence of phenothiazine, thionol and leucothionol.

In each instance where a red dye was removed from the bile, the product displayed the reactions typical of thionol. It could be easily extracted from aqueous solution by either ether or chloroform. In aqueous solution it could be easily reduced to the leuco form by stannous chloride or sodium hydrosulfite, and reoxidized with hydrogen peroxide.

These evidences for the biliary excretion of phenothiazine, thionol, and leucothionol in rabbits, dogs, and man suggest an explanation for the presence of the substances in the intestinal tract, over and above any unabsorbed phenothiazine, or thionol and leucothionol formed from phenothiazine in situ.

When considered in the light of the earlier reports (2) on the bactericidal properties of phenothiazine, it is suggested that the therapeutic value of orally administered phenothiazine be tested in cases of cholecystitis where bacterial infection is known or suspected of being present. Parasiticidal effects might be expected in the intestine, and indeed an anthelmintic action has been reported by parasitologists.

ANTHELMINTIC ACTION

Phenothiazine is remarkable in that, following gastric or oral administration, it is rapidly absorbed despite its low water solubility. A part of the phenothiazine administered is oxidized to the reversible oxidation-reduction system thionol-leucothionol. At present it is not known whether this oxidation takes place in the digestive tract or in the tissues following absorption of phenothiazine. In any event there is excreted in the urine (8) and in the bile phenothiazine, thionol and leucothionol.

In the urine, at least, there is also leucothionol in some conjugated form from which it is liberated easily by acid hydrolysis. The exact manner of conjugation is not understood at present, nor is it known how the phenothiazine is kept in solution in bile and urine. When precipitated from these media by the addition of hydrochloric acid, phenothiazine cannot be brought back into solution by neutralization with sodium hydroxide. Even if phenothiazine is completely absorbed, its presence together with thionol and leucothionol in the bile would explain the presence of all three compounds in the intestinal tract.

The conversion of phenothiazine to thionol takes place readily in the

presence of air and moisture and, in the presence of appropriate reducing agents, variable amounts of thionol are reduced to leucothionol. If shaken with water to form a saturated solution, phenothiazine will, in the course of two or three days, be converted to thionol and confer a red color to the solution. Chemically, thionol is closely related to the other two thiazine dyes, Lauth's violet and methylene blue, and like them is a reversible oxidation-reduction system. This system has been studied potentiometrically by DeEds and Eddy (10) and found to differ from the other two thiazines by being much more easily reduced. Its potential places it much closer to the oxygen electrode than either Lauth's violet or methylene blue. At pH 7 the E_o' for thionol is 0.158 volts, or about midway between 1-napthol-2-sulfonate-indophenol and 2, 6-dichlorophenol-indo-ortho-cresol.

This ready conversion of phenothiazine to thionol and the position of the latter on the oxidation-reduction scale may have an important bearing on the mechanism of action as a bactericidal agent, insecticide, or as an anthelmintic. All living things have one characteristic in common, namely, respiratory processes, whether they be aerobic or anaerobic. The presence of a strongly positive oxidation-reduction system in adequate amount could conceivably interfere with the fundamental respiratory mechanisms. Thionol-leucothionol occupies a position on the oxidation-reduction scale much more positive than the natural oxygen carriers, cytochrome, pyocyanine, flavoprotein, ascorbic acid, flavine phosphate, and lactoflavine; and more positive than such reversible enzyme systems as succinic-fumaric, lactate-pyruvate, and xanthine-uric acid.

The remainder of this paper presents the evidence obtained from experiments in vitro to show that thionol has a definite effect on hog ascaris (*Ascaris lumbricoides*) whereas phenothiazine has no action as compared with an appropriate control. As stated above, the administration of phenothiazine leads to the biliary excretion of phenothiazine, thionol, and leucothionol which subsequently pass into the intestinal tract. Thionol is therefore present in the intestine and available for an anthelmintic action if such exists.

Hog ascaris and a sample of hog bile were obtained from a local slaughter house, taken to the laboratory as rapidly as possible, and kept in an incubator at 37° C until ready for use. A series of five conical-shaped urine sedimentation glasses were placed in a water bath maintained at a temperature of 37°–39° C. The basic medium employed in each vessel was 0.9 per cent sodium chloride. The first control medium was prepared by adding 10 cc of chloroform to 100 cc of saline solution, boiling to expel the chloroform, and making up to 100 cc with distilled water. This was done so that the saline control would contain traces of chloroform, if any, comparable to the amounts remaining in the vessels

containing the saturated phenothiazine and thionol solutions. The second control medium was the same as the first, except that 5 cc of hog bile were added. A saturated phenothiazine solution was prepared by adding 10 cc of a chloroform solution of phenothiazine to 100 cc of saline solution, boiling to expel the chloroform, filtering while hot, and making up to 100 cc with distilled water. A saturated thionol solution was made in a similar manner. The fifth vessel contained a saturated thionol solution prepared in the same way, but with the addition of 5 cc of hog bile. When the temperature of the 100 cc medium in each vessel reached equilibrium with that of the water bath, two active worms were placed in each vessel. This experiment was performed five times, the only variable being the amount of bile added to the fifth vessel. The periods of observation varied from two to six hours. The results of a typical experiment are given in Table 1.

The results obtained were the same and surprisingly clear-cut in all the tests made. The worms in both controls and in the phenothiazine solution remained active throughout the periods of observation. The worms placed in the thionol solution exhibited increased activity, and in some instances attempted to crawl up the sides of the vessel. This increased activity, indicative of irritation, was followed by depression and complete inactivity. As the experiment progressed there was, however, some return of activity which tended to increase as the intensity of color of the thionol solution decreased, a phenomenon to be discussed presently. The worms placed in the thionol solution to which 5 cc of bile had been added became depressed and inactive very early, showing little if any evidence of a preliminary stimulation or irritation. In other words, the presence of bile shortened or eliminated the first stage of action and facilitated development of the depressant action. However, the tendency to regain activity was present as in the thionol solution, but occurred earlier, and the disappearance of the characteristic red color of thionol took place sooner.

The disappearance of color in the thionol and thionol plus bile solutions may be considered in conjunction with the observed resumption of activity by the worms. In the case of thionol alone, the worms became slightly red in color, while the medium was losing color. The loss in color of the medium was undoubtedly due, in part, to a taking up of thionol by the worms, but was due in large measure to a reduction of thionol to leucothionol. Proof of the existence of leucothionol in the medium was obtained by adding hydrogen peroxide to a few cc of the bleached medium and obtaining an intensification of color. As stated above, the worms placed in a thionol solution containing 5 cc of bile showed an earlier onset of depression, and an earlier return of activity, which coincided with a more rapid disappearance of the thionol color.

TABLE 1.—*Comparative effects of phenothiazine and thionol on* Ascaris lumbricoides

Time	Control 1 0.9 per cent sodium chloride	Control 2 0.9 per cent sodium chloride plus chloroform (see text)	0.9 per cent sodium chloride saturated with phenothiazine	0.9 per cent sodium chloride plus 0.01 per cent thionol	0.9 per cent sodium chloride plus 0.01 per cent thionol plus 5 cc bile
10 : 46	Active	Active	Active	Activity greater than controls	Least active
10 : 51	"	"	Few escape efforts	Many escape efforts	Activity increasing
11 : 00	"	"	Active	Escape efforts	Active but less so than controls
11 : 15	"	"	"	Little activity	Least active
11 : 30	"	"	"	" "	Inactive
11 : 45	"	"	"	" "	"
12 : 00	"	"	"	" "	"
12 : 30	"	"	"	Very little activity	Slight activity
12 : 50	"	"	"	No activity	"
1 : 10	"				

Since the control containing bile had no depressant action on the worms, the effect of bile in the presence of thionol must have been due to some factor such as the lowering of surface tension which might facilitate penetration of the dye into the tissues. Since bile itself causes some reduction of thionol to leucothionol, thereby lowering the concentration of effective thionol, and since the worms tend to recover their activity when the thionol is reduced, the reason for the more rapid recovery of the worms in thionol plus bile is evident.

Since bile facilitates the depressing action of thionol, but later hastens the return of activity, smaller amounts of bile might preserve the synergistic effect on the depressant action, but not convert sufficient thionol to leucothionol to facilitate recovery. Therefore additional experiments employing thionol plus bile were performed using 1 cc, 0.5 cc, 0.1 cc, and 0.05 cc of bile. In the vessel containing thionol plus 0.05 cc of bile the worms behaved about the same as in a medium of thionol alone. The other concentrations definitely hastened the depressant action of thionol, but did not cause sufficient reduction to leucothionol to appreciably shorten the period of depression. In other words, in the presence of a given amount of thionol there was an optimum concentration of bile which produced depression of the worms without shortening the period of recovery.

These in vitro results leave no doubt that, under the conditions of the experiment, thionol and not phenothiazine depresses the activity of hog ascaris, and that the effect may be augmented by an appropriate concentration of bile. The transfer of information obtained in vitro on one species of parasite to any explanation of anthelmintic action, in general, is recognized as lacking a certain validity. However, it must be granted that in vitro experiments do permit a control of conditions, and an examination of factors difficult to study in the intact animal. Finally, much of the value of such data depends upon the interpretation.

The evidence at hand shows that thionol, as well as leucothionol and phenothiazine, are present in the intestinal tract following administration of phenothiazine due to excretion in the bile. The depressant action of thionol plus bile observed in vitro was of limited duration, the time being determined by the time required to reduce the thionol to leucothionol. If conditions would permit the reoxidation of leucothionol to thionol, or if the concentration of thionol were maintained by the addition of a fresh supply the depressant action would likely persist. These theoretically more favorable conditions exist in the intact animal. The administration of phenothiazine may be repeated, and even with a single dose there tends to be a prolongation of action as the biliary excretion of thionol rises to a peak and then subsides relatively slowly. Meanwhile intestinal peristalsis would tend to remove the parasites before recovery is complete. The

evidence presented is, at least, highly suggestive that the anthelmintic action of phenothiazine is due to the thionol excreted into the intestine by way of the biliary tract and that bile facilitates this action.

MECHANISM OF ACTION

It is both desirable and fortunate that this effective anthelmintic exhibits little or no toxicity to the host. Although the argument may be regarded as highly speculative it is appropriate to make a suggestion as to the mechanism responsible for an effective toxic action toward bacteria, insects, and intestinal parasites, whilst mammals, dependent upon the hemoglobin system for oxygen transport, show few if any toxic symptoms. A detailed discussion of a probable mechanism would lead far afield into the complicated subject of biological oxidation, quite outside the scope of the present paper, but a suggestive hypothesis may be briefly stated.

At a pH of 7 the potential of the hemoglobin-methemoglobin system is + 0.152 volts. At the same pH the potential of the thionol-leucothionol system is + 0.158 volts. This slightly more positive potential of the thionol-leucothionol system is not likely to cause an appreciable shift of the hemoglobin-methemoglobin system toward methemoglobin and interfere with the oxygen transport mechanism characteristic of hemoglobin-oxyhemoglobin. In other biological systems, however, where oxygen transport is dependent upon such natural oxygen carriers as cytochrome, pyocyanine, flavoprotein, ascorbic acid, flavine phosphate, and lactoflavine, a different situation prevails. These systems are all much more negative in potential, could easily reduce thionol to leucothionol, and in turn be oxidized. Especially under conditions where a fresh supply of thionol is being supplied by oxidation of phenothiazine, as in the use of phenothiazine for anthelmintic purposes, for treatment of cystitis of bacterial origin, and in the slow but persistent oxidation of phenothiazine used as an insecticidal spray, it is conceivable that the natural oxygen carriers might be kept in an oxidized state, unable to function, and thus cause the depression and ultimate death of the organism.

SUMMARY

1. Gastric or oral administration of phenothiazine has been shown in one man, two rabbits, and three dogs to result in the biliary excretion of phenothiazine, thionol, and leucothionol.

2. In vitro phenothiazine had no demonstrable action on hog ascaris.

3. Thionol first caused stimulation of hog ascaris, followed by depression which persisted as long as an adequate concentration of thionol was present.

4. Bile facilitated the development of the depressant action of thionol on hog ascaris.

5. An hypothesis for the mechanism of thionol action has been briefly outlined.

Bibliography

(10) DeEds, F. and Eddy, C. W. 1938 Studies on phenothiazine: IV. Potentiometric characterization of thionol. J. Am. Chem. Soc. **60**: 2079.

(8) DeEds, F., Eddy, C. W. and Thomas, J. O. 1938 Studies on phenothiazine: V. Fate of phenothiazine in the body. J. Pharmacol. and Exper. Therap. **64**: 250.

(2) DeEds, F., Stockton, A. B. and Thomas, J. O. 1939 Studies on phenothiazine: VIII. Antiseptic value of phenothiazine in urinary tract infections. J. Pharmacol. and Exper. Therap. **65**: 353.

(3) Gordon, J. McL. 1939 A preliminary note on the chemotherapy of oesophagostomiasis of sheep with special reference to the efficiency of phenothiazine. J. Council Scient. and Indust. Research, Australia **12**: 203. (Chem. Abs. 1940 **34**: 159.)

(4) Gordon, J. McL. and Whitten, L. K. 1939 A preliminary note on the anthelmintic efficiency of phenothiazine against *Haemonchus contortus* in sheep. J. Council Scient. and Indust. Research, Australia **12**: 207. (Chem. Abs. 1940 **34**: 160.)

(6) Harwood, P. D., Jerstad, A. C. and Swanson, L. E. 1938 The efficacy of phenothiazine for the removal of ascarids and nodular worms from swine. J. Parasitol. **24** Suppl: 16.

(5) Roberts, F. H. S. 1939 A preliminary note on the treatment of oesophagostomiasis in sheep with phenothiazine. J. Council Scient. and Indust. Research, Australia **12**: 208. (Chem. Abs. 1940 **34**: 160.)

(1) Smith, L. E., Munger, F. and Siegler, E. H. 1935 Phenothiazine, a promising new insecticide. J. Econom. Entom. **28**: 727.

(7) Swales, W. E. 1939 Tests of phenothiazine. A highly efficient anthelmintic. Canad. J. Comp. Med., July issue.

(9) Thomas, J. O., DeEds, F. and Eddy, C. W. 1938 Studies on phenothiazine: VII. The bactericidal properties of urine after oral administration of phenothiazine. J. Pharmacol. and Exper. Therap. **64**: 280.

DIPLORCHIS SCAPHIOPI, A NEW POLYSTOMATID MONOGENEAN FLUKE FROM THE SPADEFOOT TOAD*

L. Ormand Rodgers

Twenty of 62 spadefoot toads, *Scaphiopus bombifrons* Cope, collected near Stillwater, Oklahoma, in April and June 1940, were found to harbor 45 specimens of an apparently new ovoviviparous polystomatid fluke of the genus *Diplorchis* Ozaki, 1931. The parasites were found in the urinary bladder, 5 being the maximum number from a single host. The worms obtained in April were all mature and contained fully developed embryos. Indeed, eggs with delicate external membranes were deposited in large numbers in water in which the worms were collected. These quickly hatched, producing active, externally ciliated, tetraoculate larvae. The eyespots and moving cilia of larvae in utero could be seen in adults under low magnification. Some of the worms collected in June were small and immature; in these the reproductive organs were only partially differentiated and eyespots, in various stages of disintegration, were still discernible. Mature flukes taken in June contained and deposited fewer embryonated eggs than those of April.

The trematode was studied both alive and preserved. Fixatives used were Bouin's and acetic-sublimate, the stain borax-carmine. Measurements were taken from preserved specimens which had not been subjected to pressure at the time of killing. Mechanically flattened animals, however, were much more useful for the study of anatomical details.

Diplorchis scaphiopi n. sp.

(Figs. 1–5)

Description: Measurements (length × width, unless otherwise stated) from 14–17 preserved and mounted adult specimens of the April collection. Total length 3.39 (2.15–4.35) mm. Body proper 2.45 (1.49–3.12) × 1.31 (0.78–1.78) mm. Haptor 763 (535–959) × 943 (666–1233) μ. Oral sucker 173 (140–219) × 228 (180–280) μ. Oral aperture 95 (73–126) μ wide. Pharynx 163 (112–213) × 110 (58–166) μ. Cirrus pouch 46 (40–52) μ wide. Ovary 209 (133–466) × 125 (60–286) μ. Testes too vaguely outlined and difficult to locate in adult unflattened specimens for measurement. Macrohooks 265 (179–406) μ long. Microhooks 23 (18–28.5) μ long. Acetabula 245 (159–326) μ in diameter.

Body proper lanceolate, somewhat pointed at anterior end. Haptor subhexagonal and provided ventrally with 6 prominent acetabula, 16 microhooks, and 2 macrohooks. Positions of vulvae represented by paired marginal prominences at about one-third length of body proper from front end. Integument covered everywhere with a pattern of minute, contiguous, polygonal areas readily visible only under oil immersion objective; skin also densely beset with minute short papillae. In subadult animals the 2 tandem pairs of black-pigmented embryonic

Received for publication, July 2, 1940.

* Contribution No. 75 from the Zoological Laboratory, Oklahoma Agricultural and Mechanical College, prepared under the direction of R. Chester Hughes.

eyespots may still be seen, located dorsal to the front end of the pharynx, the front spots being slightly the closer together. Eyespots were not observed in larger specimens. Microhooks located, 1 in each acetabulum, 6 in a transverse row between the anterior acetabula, and 4 in a row near posterior margin between the back suckers. Macrohooks compressed, close together, with their broad, longitudinally striated, bases near center of haptor and their ventrally directed points farther back.

Oral sucker terminal, mouth ventrosubterminal. Pharynx pyriform, small end forward. Ceca clearly demonstrable only in living specimens. In young animals the ceca are very broad, have forward extensions lateral to the pharynx, are confluent in the haptor, and are provided with numerous short lateral and longer medial diverticula. In older animals the latter, on opposite sides of the body, become connected by a dozen or more irregularly arranged transverse commissures, which in turn may themselves be connected by longitudinal anastomoses. Many of the parasites, when first collected, had the enteron filled with a brownish fluid presumed to be partially digested blood. Numerous pigmented cells, clearly recognized only in living specimens, occur associated with (in or on) the walls of the ceca. These cells are circular to ellipsoidal in outline, flattened parallel to the intestinal wall, and rather uniformly distributed over the enteron although widely separated from each other. In each cell the hyaline nucleus and numerous discrete jet-black granules in the cytosome were clearly visible. After preservation these chromatophores cease to "stand out" individually and the pigmentation soon fades.

Paired urinary vesicles spheroidal, dorsosubmarginal between levels of pharynx and vulvae, with dorsal pores. In live worms the bladders were seen to pulsate frequently and alternately.

Testes symmetrically paired, large and subequatorial in young specimens, relatively smaller and farther forward in older ones. Vasa efferentia extend mediosubanteriad, uniting to form long sinuous forward-running vas deferens. Cirrus pouch located ventrosubmediad behind pharynx, armed with 8 hair-like setae.

Ovary ovoidal, longitudinally elongate, located to one side (right or left in equal frequency) of median line at level between vulvae and testes. Oviduct issues directly posteriad from back end of ovary; it soon curves sharply ventroanteromediad; next it gives off the genitointestinal canal which runs laterad ventral to oviduct to join ventral side of intestinal ramus ipsilateral with ovary; next it gives off the submedian common forward-reaching vitelline duct or vitelline reservoir; thence it curves abruptly posteriad and enters Mehlis' gland. The latter, relatively larger and more diffuse in young than in older worms, occupies more or less completely the interstice between ovary and testes. Uterus makes a principal posteriad loop reaching to or into haptor; descending limb slightly sinuous and largely confined to median plane; ascending limb, in adults, thrown into numerous transverse loops that extend across almost entire width of body, dorsal to descending limb. Uterus and cirrus pouch connect with a narrow common genital atrium which opens to exterior through a submedian pore ventral to pharynx.

Vitellaria a submarginal pair of small compact bodies at level between vulvae and ovary. Paired vitelline ducts run posteromediad to join short submedian common duct extending back to oviduct. Anterior end of common vitelline duct sometimes enlarged as a yolk reservoir. Vaginae extend medioposteriad from vulvae along medial surfaces of vitellaria to join proximal ends of paired vitelline ducts.

Along length of uterus one observes eggs, surrounded by delicate membranes, in all stages of development from unicellular zygote to ready-to-hatch larva. Most of ascending limb is occupied by the latter to the number of 2 or 3 dozen.

Free-swimming larva: Measurements from several preserved specimens. Body 403 (333–446) × 174 (139–213) μ. Oral sucker 69 (61–79) × 85 (67–97) μ; mouth 32 (22–40) μ wide. Pharynx 47 (34–63) × 34 (28–42) μ; prepharynx 26 (18–39) μ long. Haptor 113 (100–153) × 146 (126–179) μ.

Body ellipsoidal, a little more pointed anteriorly and with haptor well defined but not sharply set off. Microhooks similar in shape, size and number to those of adult. Macrohooks represented by partially developed feebly chitinized, rod-like

bodies. Large ciliated epithelial cells in undetermined number and arrangement more or less cover body proper and dorsal surface of haptor. Cerebral ganglia and interconnecting supraprepharyngeal commissure clearly observed. Two pairs of large heavily pigmented eyespots, the posterior ones being a little the larger, occur in direct tandem arrangement associated with antero- and posterodorsal surfaces of ganglia. In living specimens eyespots are deep-black in transmitted light, milky white and opalescent in reflected light.

Host: Scaphiopus bombifrons Cope, 1863.
Habitat: Urinary bladder.
Locality: Stillwater, Oklahoma.
Cotype specimens: Several in toto mounts in U. S. Nat. Mus. Helm. Coll.

The genus *Diplorchis* Ozaki, 1931, contains three previously known species, namely, *D. ranae* Ozaki, 1931, from *Rana rugosa* in Japan, *D. nigromaculatus* Lee, 1936, from *Rana nigromaculata* in China, and *D. americana* Rodgers and Kuntz, 1940, from *Scaphiopus couchii* in Oklahoma. The writer cannot offer a comparison of the new species with *D. nigromaculatus* because prolonged and repeated efforts to obtain the description (Lee, 1936) of the latter were in vain.

Diplorchis scaphiopi differs from *D. ranae* in having (1) integumentary papillae; (2) the microhooks more uniform in shape and size, the 2 hindmost pairs forming a transverse row; (3) compact vitellaria; (4) 8 instead of 10 setae in the cirrus pouch; (5) ovoviviparity; (6) a scaphiopodid instead of a ranid host; (7) a New World distribution. The new species differs from *D. americana* in being (1) less than half as large with regard to most of the principal measurements—the larva is also smaller, and in having (2) integumentary papillae, (3) macrohooks —this difference true of both adult and larva, (4) 8 instead of 6 setae in the cirrus pouch, and (5) the eyespots of the larva heavily pigmented.

Diplorchis belongs to the family Polystomatidae Gamble, 1896, an important review of which is that of Price (1939).

Bibliography

Lee, L. Y. 1936 On a new and rare trematode. Contrib. Inst. Zool. Peiping **3**: 123–132.

Ozaki, Y. 1936 Studies on the frog-trematode *Diplorchis ranae*. I. Morphology of the adult form with a review of the family Polystomatidae. J. Sc. Hiroshima Univ., s. B, Div. 1, Zool. **3**: 193–225.

Price, E. W. 1939 North American monogenetic trematodes. IV. The family Polystomatidae (Polystomatoidea). Proc. Helm. Soc. Washington **6**: 80–92.

Rodgers, L. O. and Kuntz, R. E. 1940 A new polystomatid monogenean fluke from a spadefoot. Wasman Collector **4**: 37–40.

Yamaguti, S. 1936 Studies on the helminth fauna of Japan, Part 14. Amphibian trematodes. Japan. J. Zool. **6**: 551–576.

Explanation of Plate

All figures concern *Diplorchis scaphiopi* n. sp. Fig. 2 is a freehand study; the others were drawn with the aid of a camera lucida with details added freehandedly.

Abbreviations

b—excretory bladder
c—genitointestinal canal
g—enteron
m—Mehlis' gland
o—ovary
t—testis
u—uterus
v—vagina
y—vitellarium

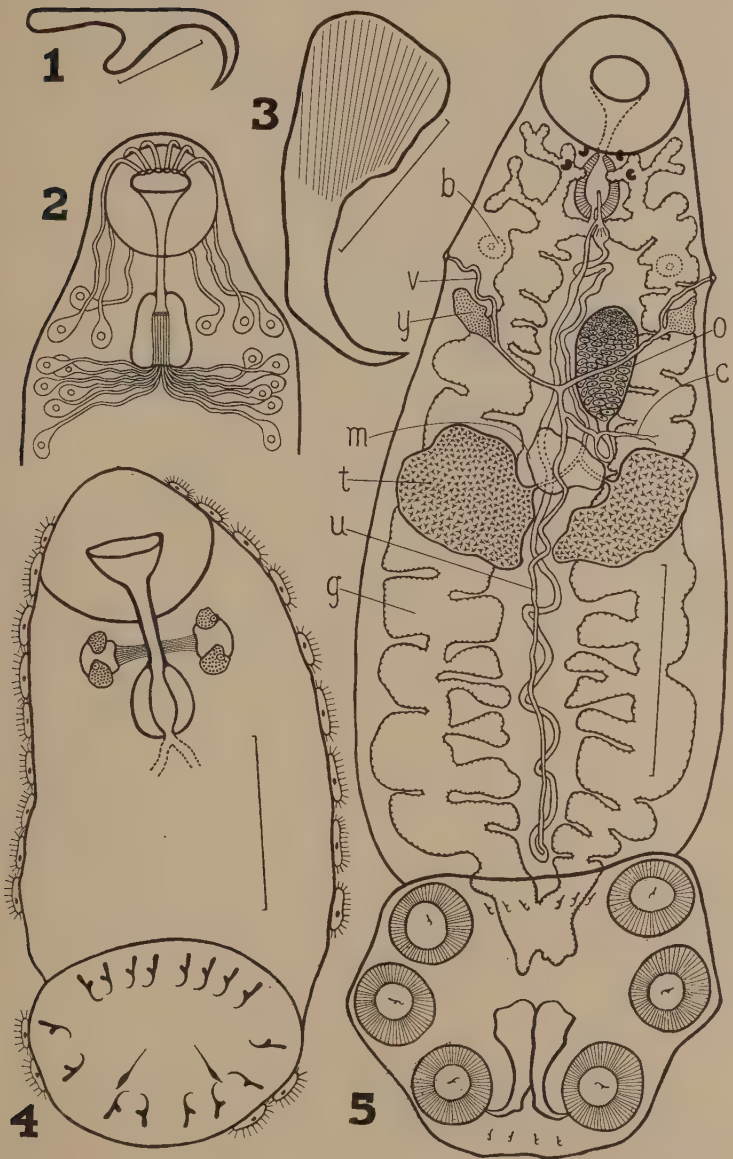
Fig. 1. Small haptoral hook from an adult. Scale, 12.5 μ.

Fig. 2. Unicellular glands about mouth and pharynx. The exact number of cells in either group was not determined.

Fig. 3. Macrohook. Scale, 100 μ.

Fig. 4. Free-swimming larva, ventral view. Scale, 100 μ.

Fig. 5. Subadult, ventral view. The uterus and enteron have not reached their maximum complexity. The ovary and testes are relatively larger and more posteriad than in older animals. Scale, 500 μ.


1
2
3
4
5
b
v
y
o
c
m
t
u
g

STUDIES ON MONOGENETIC TREMATODES. IV. *ANCHORADISCUS*, A NEW DACTYLOGYRID GENUS FROM THE BLUEGILL AND THE STUMP-KNOCKER SUNFISH[1]

JOHN D. MIZELLE

The present author has been opposed to the creation of new genera of TETRAONCHINAE on the basis of characters of questionable value. In 1938 he and Hughes reduced nine North American genera to three, because of character differences considered insufficient for separation.

During the examination of rather extensive material collected from Florida fishes and kindly sent to the author by Dr. R. V. Bangham (College of Wooster, Wooster, Ohio), a type of TETRAONCHINAE, one species of which was described by Summers (1937) as *Actinocleidus triangularis*, has been found to differ enough in the nature of the haptoral armament to warrant generic distinction. The writer feels that the characters distinguishing this proposed genus are of sufficient magnitude to stand the test in a revision of North American TETRAONCHINAE which is to come at a later date. The present author believes that the described forms of North American TETRAONCHINAE are inadequate in number for presentation of a good picture of present evolutionary radiation, expressed in morphological detail, and that until the picture has been rendered more complete, a comprehensive taxonomic revision would be untimely.

Anchoradiscus n. g.

Diagnosis: TETRAONCHINAE with anchor bases situated parallel to, and developed so as to extend through the greater portion of a frontal plane of, the discoidal haptor. Anchor shafts vestigial or wanting; anchor points (recurved) directed laterally. Haptoral bars two, attached to the ventral (lateral) surfaces of the anchor bases and articulated with each other in midportions. Each of the fourteen hooks situated on the haptoral margin and differentiated into a base, a solid slender shaft, a sickle-shaped termination, and an opposable piece. Copulatory complex consisting of a cirrus and accessory piece; vagina sinistral. Eye spots four, posterior pair larger. Parasitic on the gills of fresh-water fishes.

Type species: Anchoradiscus anchoradiscus n. sp.

Received for publication, July 6, 1940.

[1] Contribution No. 76 from the Zoological Laboratory, Oklahoma Agricultural and Mechanical College, Stillwater.

Grateful acknowledgment is hereby made to Dr. W. A. Summers of Tulane University, for loan of figures and other information concerning *Anchoradiscus triangularis* (Summers, 1937) and also to Dr. E. W. Price of the Zoological Division of the Bureau of Animal Industry (U. S. D. A.), for checking the validity of the proposed generic name, *Anchoradiscus*.

Anchoradiscus anchoradiscus n. sp.

(Figs. A–D)

Description: Medium-sized dactylogyrids with a smooth cuticula; cephalic lobes usually well defined. Length 0.692 mm (0.456–0.945 mm), greatest width 0.187 mm (0.104–0.323 mm). Eye spots four, members of the posterior pair larger and closer together than those of the anterior pair. Pharynx circular or subcircular in outline and may be broader than long or longer than broad; greatest transverse diameter 0.049 mm (0.029–0.065 mm).

Haptor discoidal in shape and of a diameter greater than the body width; greatest transverse diameter 0.313 mm (0.185–0.484 mm). Bars two, situated ventral to anchors, similar in shape, and with midportions bent toward, and articulated to, each other. Arms of anterior bar hollow and with well-defined articulating surfaces for attachment to anchors (Fig. B, 1). Posterior bar solid, articulations to anchors well defined (Fig. B, 5). A pair of knobs present on each bar at site of attachment with each other. Knobs of anterior bar apparently loosely attached to bar and each member of the pair with a strand of tissue reaching to the adjacent haptoral surface (Fig. B, 3). Proximal portion of each arm of posterior bar often with an accessory structure connecting it with the lateral portion of the haptor (Fig. B, 4). Greatest distance between arm tips of anterior bar 0.144 mm (0.100–0.216 mm); greatest distance between arm tips of posterior bar 0.137 mm (0.085–0.167 mm). Anchors thin, similar in shape, anterior pair larger; bases enormously developed, occupying the major portion of a frontal plane of the haptor, and limiting the greater portion of the haptoral outline (Fig. A, 8 and 11). Anchor shafts reduced in length and parts (former) of them apparently incorporated in the expanded anchor bases and each evident as a reinforced marginal portion of the base (Fig. D, 5). Separation of bases into roots barely indicated or absent in some specimens (Figs. A, B, D). Length of anterior anchors 0.179 mm (0.103–0.219 mm), greatest width 0.110 mm (0.063–0.144 mm); length of posterior anchors 0.160 mm (0.100–0.190 mm), greatest width 0.103 mm (0.076–0.140). Anchor wings well developed. Hooks similar in size and shape, fourteen in number, and situated around the margin of the haptor (Fig. A). Each hook consists of an oval base, a slender solid shaft, a sickle-shaped termination, and an opposable piece. Hook bases longitudinally ovate in outline and approximately the same length as the shaft. Hook lengths 0.017–0.021 mm.

Gonads ovoid in outline and located in rear half of body proper. Testis situated dorsoposterior to, and larger than, the ovary. Vagina on left body margin near the midportion. Copulatory complex consisting of a cirrus and accessory piece. Cirrus with a large base and a curved tapered terminal portion (Fig. C, 2). Accessory piece consisting of a thin expanded basal portion, which partially encloses the basal portion of the cirrus, and a solid recurved termination (Fig. C, 1). Cirrus length (distance between tip and most distal part of the base) 0.031 mm (0.024–0.039 mm); accessory piece approximately the same length as the cirrus. Vitellaria well developed and with arrangement as in the genus *Urocleidus* Mueller, 1934. The nature of the gut could not be determined from fixed material.

Type hosts and localities: Stump-knocker sunfish, *Eupomotis microlophus* (Günther, 1859) Englewood Ditch, Englewood, Fla.; Myakka River State Park, East Sarasota, Fla.; Lake Okeechobee, Moore Haven, Fla.; and Everglades Canal, Naples, Fla. Bluegill sunfish, *Lepomis macrochirus* Rafinesque, 1819, Canal, North Everglades, Fla.

Site of infestation: Gills.

Number of specimens used in this description: Twelve.

Type specimens: Cotypes Nos. 36773–6 U. S. Nat. Mus., Washington, D. C.

In 1937 Summers described *Anchoradiscus triangularis* from the gills of *Lepomis symmetricus* Forbes, 1883, and assigned it to the genus *Actinocleidus* Mueller, 1937. Although specimens of this parasite were

not available for comparison, the writer secured several figures and notations from Dr. Summers and these together with the published description have shown this form to be a close relative of *Anchoradiscus anchoradiscus* and should be placed in the newly proposed genus.

The general morphology of species of *Anchoradiscus* is very similar to that of *Actinocleidus* forms. In addition, the haptor is discoidal and the bars are articulate in both genera. Differences between species of the genera are manifest in that those of *Anchoradiscus* possess (1) a thinner haptor, (2) anchors which lie parallel to the haptoral surfaces (dorsoventral) with their recurved points projecting laterally instead of ventrally as in *Actinocleidus* species, (3) bars which lie ventral to the anchors and attach to the lower lateral anchor surfaces instead of being located between the two pairs of anchors and attaching to them between the roots of the anchor bases as in *Actinocleidus* forms,[2] (4) hooks with large ovate bases instead of small spherical-to-ovate ones as in members of *Actinocleidus*, (5) anchor shafts which are much reduced and may be interpreted as wanting since the edge of the anchor base, which is in line with the vestigial shaft, is comparatively thick and probably includes a part or all of the anchor shaft, and (6) enormously developed anchor bases which when taken together (all four), occupy most of a frontal plane of the haptor. This last point presents a sharp contrast with reference to the relatively small anchor bases in species belonging to other genera of North American fresh-water TETRAONCHINAE.

It is very probable that the gut of *A. anchoradiscus* is bifurcate and posteriorly confluent as described for its closest relative (*A. triangularis*) by Summers (1937).

BIBLIOGRAPHY

MIZELLE, J. D. AND R. C. HUGHES 1938 The North American fresh-water Tetraonchinae. Am. Midland Naturalist **20**: 341–353.

MUELLER, J. F. 1937 Further studies on North American Gyrodactyloidea. Ibid. **18**: 207–219.

SUMMERS, W. A. 1937 A new species of Tetraonchinae from *Lepomis symmetricus*. J. Parasitol. **23**: 432–434.

[2] Attachment of the bars to the anchor bases has not been definitely established for all species of *Actinocleidus*. Attachment is thought to be made between the roots of the anchor bases or in this region in cases where the roots are absent.

Explanation of Plate

All drawings were made with the aid of a camera lucida at the designated magnifications.

Fig. A (ventral view)

1. Head organs
2. Eye spots
3. Pharynx
4. Copulatory complex
5. Vagina
6. Ovary
7. Testis
8. Anterior anchor
9. Anterior bar
10. Posterior bar
11. Posterior anchor
12–15. Hooks

Fig. B

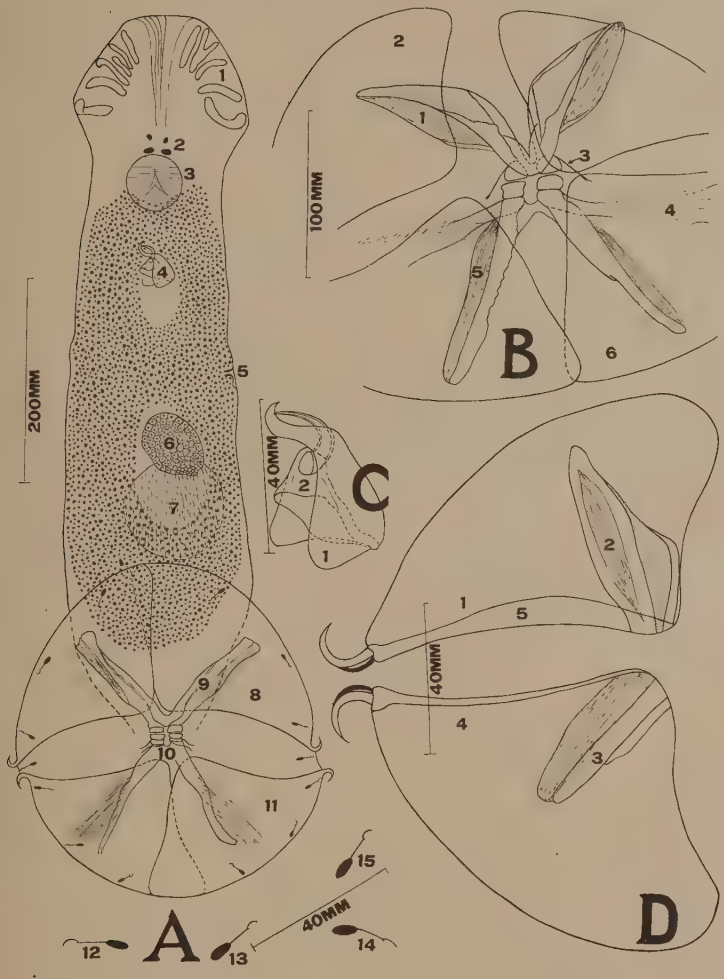
1. Anterior bar
2. Anterior anchor
3. Strand of tissue from knob
4. Accessory bar-structure
5. Posterior bar
6. Posterior anchor

Fig. C

1. Accessory piece
2. Cirrus

Fig. D

1. Anterior anchor
2. Part of anterior bar
3. Part of posterior bar
4. Posterior anchor
5. Thickened portion of anchor base


1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
200MM
40MM
A
100MM
B
C
D

DENDRITOBILHARZIA ANATINARUM N. SP., A BLOOD FLUKE FROM THE MALLARD[1]

E. L. Cheatum[2]

A new blood fluke of ducks belonging to the genus *Dendritobilharzia* Skrjabin, 1920, is here described, with a discussion on the occurrence of blood fluke eggs and their possible pathological significance to ducks.

Subfamily Bilharziellinae Price, 1929

Price (1929) in his synopsis of the Schistosomatidae proposed the subfamily Bilharziellinae to include the genera *Bilharziella* Looss, 1899, *Gigantobilharzia* Odhner, 1910, *Trichobilharzia* and *Dendritobilharzia* Skrjabin and Zakharow, 1920. Ejsmont (1929) added the genus *Pseudobilharziella*. The diagnosis given by Price for this subfamily includes a statement regarding character of the uterus, describing it as short and containing a single egg. This would exclude the species described here since it is long, coiled, and contains many eggs. All other structural characters agree with this subfamily diagnosis, and since the character of the uterus alone should not constitute a subfamily difference, the diagnosis is accordingly amended to read:

Bilharziellinae Price, 1929, emend.

Schistosomatidae. Male and female similar in form, either flattened or thread-like. Suckers present or absent. Gynaecophoric canal absent or imperfectly formed. Paired intestinal ceca short, common cecum long, with or without lateral dendritic branches. Testes numerous, situated along course of common cecum. *Uterus may contain one or many eggs.*

Genus *Dendritobilharzia* Skrjabin, 1920

In examining the genera of Bilharziellinae, the characters of *Dendritobilharzia* come most nearly corresponding to the form described here. The differences in character of the uterus, the number of eggs contained, and larger size of the female as compared to the male are not considered of sufficient significance to warrant establishing a new genus, therefore the generic diagnosis is amended to read:

Dendritobilharzia Skrjabin, 1920, emend.

Bilharziellinae. Body of both sexes elongate and flattened. *Female larger or smaller than male.* Cuticle without spines or tubercules. Suckers absent. Digestive

Received for publication, July 20, 1940.

[1] Contribution from the Game Research Center, Bureau of Game, N. Y. S. Conservation Department.

[2] I wish to express my appreciation to Prof. George R. La Rue of the University of Michigan and to Dr. E. W. Price of the Bureau of Animal Industry for their kind advices, and to Dirck Benson and Douglas Beyea of the N. Y. S. Bureau of Game for identification of the ducks.

system similar to that in *Bilharziella,* common cecum long, zig-zag, provided with short, club-shaped or branched lateral ceca. Genital pore of male lateral, in anterior part of body. *Cirrus present.* Testes numerous, situated along common course of cecum, and extending from cecal union to end of body. Ovary spiral, between intestinal crura. *Uterus may contain one or many eggs.* Vitelline follicles numerous, along course of common cecum.

This genus contains two species, *D. pulverulenta,* first described by Braun (1901) as *Bilharziella pulverulenta* from *Anas platyrhynchos* collected from Africa, and *D. loossi* Skrjabin, 1924, from *Pelecanus onocrotalus* collected in the U.S.S.R. Skrjabin and Zakharow (1920) established the genus *Dendritobilharzia* for a new species of blood fluke from *Querquedula querquedula* to which they gave the name *Dendritobilharzia odhneri.* Later Skrjabin (1924) recognized *D. odhneri* as synonymous with Braun's *B. pulverulenta.* Price (1929) synonomyzed *D. odhneri* with *B. pulverulenta* and recognized *Dendritobilharzia* as the valid genus. Thus *Bilharziella pulverulenta* became *Dendritobilharzia pulverulenta* (Braun, 1901) Skrjabin, 1924. I have been unable to find reference to additional species of this genus. Brackett (1940) in listing the adult schistosomes from hosts which also occur in Wisconsin gives *Dendritobilharzia* sp. from *Melanita deglandi.* This record was obtained by personal communication with L. R. Penner who apparently has not yet described this form.

The description of this genus has been based on a striking paucity of specimens. Braun described *D. pulverulenta* from a single male worm. In his description he did not mention the presence of a cirrus although his figure shows structure indicating its presence. Semenov (1927) claims to have found the female of Braun's *pulverulenta* and described it from a single specimen. Skrjabin described *D. loossi* from a single female worm, the male remaining unknown. Thus we have only one description of the male in this genus, that of *D. pulverulenta.* There is a possibility that Braun overlooked the presence of the cirrus in writing his description, and in amending the generic diagnosis I have included the presence of a cirrus to conform with the species described here and Braun's figure of *D. pulverulenta.*

Dendritobilharzia anatinarum n. sp.

(Figs. 1, 2)

Two female and five male worms have been removed from the dorsal aortae of two mallard ducks, *Anas platyrhynchos platyrhynchos* Linnaeus. These birds were hatched and reared at the Game Research Center of the New York State Bureau of Game, located near Delmar, N. Y.

Male. Body similar in shape to female but smaller, 6.2 mm long by 0.60–0.75 mm wide; esophagus 0.427 mm long, intestinal crura united posteriorly 1.08 mm from esophageal bifurcation; testes from 120–130 in number, extending from union of intestinal crura to posterior end of body; vesicula seminalis elongate, arranged roughly in a V-shape, about 0.26 mm long, median between intestinal crura; genital pore dextral, 1.20 mm from anterior end of body; cirrus well developed, distal end bulbous.

Female. Body flattened, elongate, 8.0 mm long by 0.89–1.01 mm wide; suckers absent; oral opening sub-terminal, esophagus 0.621 mm long, intestinal crura united posteriorly 1.55 mm from esophageal bifurcation, common cecum with lateral dendritic branches, proceeding in zig-zag course to posterior end of body; ovary long, tubular, arranged in close compact loops, between intestinal crura, length and width of mass about equal, 0.525 mm; receptaculum seminis immediately posterior to ovary and median, 0.078 mm by 0.272 mm; uterus in close compact loops filling anterior half of space between intestinal crura, then proceeding anteriorly in irregular course, right of esophagus to median genital pore, 0.350 mm from anterior end of body; unpaired vitelline duct proceeding anteriorly left of ovary to oötype at level of anterior end of ovary, vitellaria filling body along course of common cecum; eggs numerous, spherical, 0.060–0.065 mm diameter, with thin, elastic, transparent shells without spines, and undeveloped when deposited (Fig. 3).

Host: Anas platyrhynchos.

Location: Dorsal aorta.

Locality: Delmar, New York.

Type: U. S. N. M. Helm. Coll. No. 39895.

The species *D. anatinarum* differs from the 2 other described species in the following main characters: 1. Uterus long, contains many eggs. The other species have short uteri with single eggs. 2. The female is larger than the male, whereas in the other species where both sexes are known the male is the larger.

DISTRIBUTION AND PATHOLOGY

During the past winter (1939–40) numerous ducks have been received at the Laboratory of Wildlife Pathology for autopsy. Many of these were found sick or dead on the State Game Farms or in the wild. Malnutrition as a result of hard winter conditions was at first suspected. Empty stomachs, emaciation, atrophic livers, blood-stained serous exudate in the body cavity, and enteritis were the usual findings. These were not with the exception of enteritis, incompatible with the general picture of malnutrition. Scrapings from intestinal linings revealed the presence of numerous spindle-shaped eggs (Fig. 4) in the mucosa and sub-mucosa. Pieces of liver and kidney pressed between two slides revealed more of the same type of eggs. Later blood flukes were found in the dorsal aorta of a mallard dying from a severe enteritis involving the lower intestine. Scrapings from the inflamed mucosa showed numerous spindle-eggs and great numbers of spherical eggs as were found being deposited by the worm described here.

Following the association between duck mortalities and the presence of blood fluke eggs, all ducks coming to the laboratory were checked for the presence of blood flukes. Microscopic examination of scrapings from the rectal and cecal mucosae for the presence of eggs was relied upon for the incidence of flukes. Table 1 shows this incidence.

It will be noted that the incidence of blood flukes is much higher in the black ducks reared on Game Farms than in those collected from the wild. This is to be expected since the constant use of game farm ponds

over a period of years and continuous habitation throughout the seasons would tend to concentrate the incidence of infection in the intermediate snail hosts. I haven't sufficient mallards from the wild to make similar comparison but the high incidence of blood flukes in the game farm birds is comparable to that of the black ducks. Incidence in the seven other species comes from birds found dead or so weak they were unable to escape capture. This selective collecting may preclude fair judgment on the actual incidence in the wild, therefore the high incidence among the canvas-back ducks may be misleading.

TABLE 1*

	Species	No. exam.	Eggs pres.
Game Farm	*Anas p. platyrhynchos*	23	18 (2)
Wild	" " "	1	1
Game Farm	*Anas rubripes*	9	7 (1)
Wild	" "	25	4
"	*Spatula clypeata*	2	1
"	*Dafila acuta*	1	1
"	*Nettion carolinense*	1	1
"	*Nyroca collaris*	1	1
"	" *valisineria*	6	5 (1)
"	" *marila*	1	1
"	*Glaucionetta clangula*	2	1
	Totals	72	41 (4)

* Data taken from specimens from Jan. 18 to June 10, 1940.
All of the forty-one (41) positive cases showed spindle eggs, and in four (4) instances, indicated in parentheses, the spherical type was also present.

The life histories of these blood flukes are being investigated at the Game Research Center preliminary to the problem of determining their pathological significance to ducks. There is good evidence at present that the eggs of *D. anatinarum* when working through the intestinal mucosa in large numbers cause hyperemia, petechial hemorrhages and may result in necrotic degeneration of the epithelium, sub-mucosa and circular muscle layer. This necrosis may result from bacterial invasion of the lesions caused by the fluke eggs. Experiments are planned to determine more accurately the role of blood flukes in producing this enteritis.

SUMMARY

A blood fluke, *Dendritobilharzia anatinarum* n. sp. is described from the common mallard, *Anas platyrhynchos* Linnaeus, with a modification of the subfamily diagnosis of BILHARZIELLINAE and a modification of the generic diagnosis for *Dendritobilharzia*. A table on the incidence of blood fluke eggs in nine species of ducks, including both the diving and surface-feeding groups, is given along with remarks on the lesions associated with their occurrence.

BIBLIOGRAPHY

BRACKETT, STERLING 1940 Studies on schistosome dermatitis. V. Prevalence in Wisconsin. Am. J. Hyg. 31 (Sec. D) : 49–63.

BRAUN, M. 1901 Zur Revision der Trematoden der Vogel. II Centr. Bakt. Abt. I. 29 : 941–948.

EJSMONT, L. 1929 Über zwei Schistosomatidengattungen der Vogel. Acad. Polonaise Sc. Math. et Nat. ser. B: 389–403.

GOWER, W. CARL 1938 Studies on the trematode parasites of ducks in Michigan with special reference to the mallard. Mem. Agric. Exper. Sta. Michigan State Coll. **3**: 1–91.

LOOSS, A. 1899 Weitere Beiträge zur Kenntnis der Trematoden-Fauna Aegyptens, zugleich Versuch einer natürlichen Gleiderung des Genus *Distomum* Retzius. Zool. Jahrb. Syst. **12**: 521–784.

MCLEOD, J. A. 1937 Two new schistosomid trematodes from water birds. J. Parasitol. **23**: 456–466.

OISO, TOMOAKI 1927 On a new species of avian schistosome developing in the portal vein of the duck, and investigation of its life-history. Taiwan Igakkwai Zasshi. September.

PRICE, E. W. 1929 A synopsis of the trematode family Schistosomidae with description of new genera and species. Proc. U. S. Nat. Mus. **75**: 1–39.

PRICE, HELEN F. 1931 Life-history of Schistosomatium douthitti (Cort). Am. J. Hyg. **13**: 685–727.

SEMENOV, V. D. 1927 Trematody ptits zapadnogo kraia S.S.S.R. (Vogel-Trematoden des westlichen Bereiches der Union S.S.R.) Sborn. rabot po gel'mintol. posv. K. I. Skrjabin (etc). Moskva, pp. 221–271.

SKRJABIN, K. I. AND ZAKHAROW, N. P. 1920 Dva novykh roda trematod iz krovi ptits (Materialy k poznaniiu gelmintofauny ptits Rossi) Zwei neue Trematodengattungen aus der Blutgefassen der Vogel. (Beitrag zur Kenntniss der Helminthenfauna der Vogel Russlands). Zzviest. Donsk. Vet. Inst. **2**: 1–5. German summary 5–6.

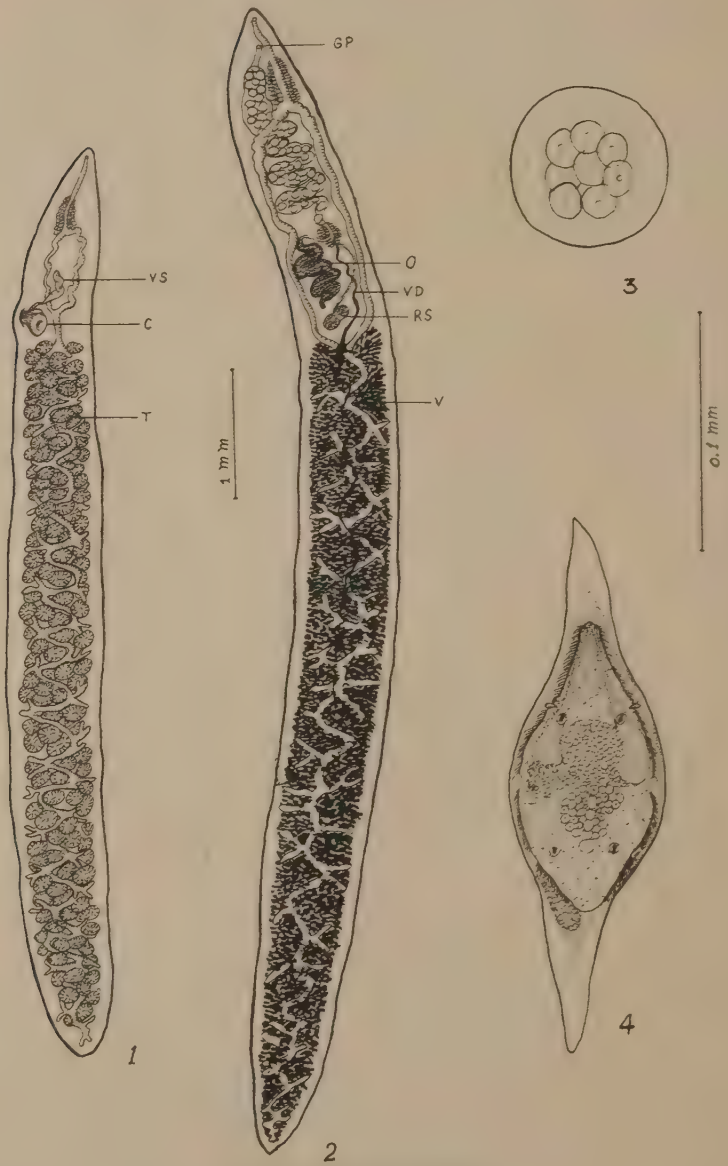
EXPLANATION OF PLATE, p. 170

FIG. 1. *Dendritobilharzia anatinarum,* adult male, ventral aspect: vs, vesicula seminalis; c, cirrus; t, testes.

FIG. 2. Adult female, ventral aspect: gp, genital pore; o, ovary; vd, vitelline duct; rs, receptaculum seminis; v, vitellaria.

FIG. 3. Egg when deposited. Spherical type.

FIG. 4. Unidentified egg of spindle type with well developed miracidium.



Dendritobilharzia anatinarum

GRYPORHYNCHUS TETRORCHIS, A NEW DILEPIDID CESTODE FROM THE GREAT BLUE HERON*

WILLIAM C. HILL

A great blue heron, *Ardea herodias herodias* Linnaeus, collected near Cromwell, Oklahoma, during the summer of 1938 was found to harbor several specimens of an apparently new tapeworm described herein as *Gryporhynchus tetrorchis*.

The worms were fixed in hot Bouin's fluid and stained with borax carmine. All studies and measurements were made on mounted specimens.

Gryporhynchus tetrorchis n. sp.
(Figs. 1–4)

Description: Largest strobila having a scolex, 15.8 mm long, with 240 proglottids—none of them gravid. Largest headless fragment, 22 mm long, with 195 segments—some of them partially gravid. Strobila slender, subcylindric, with moderately serrate margins. Proglottids all wider than long; the mature ones trapezoidal, about 130 μ long, 220 μ wide anteriorly and 273 μ posteriorly; gravid segments similar in shape to, and scarcely larger than, mature ones. Genital primordia first discernible at about 200 μ, in segment No. 38, and mature at 10.6 mm, in No. 218, from front end.

Scolex 200–253 μ long by 213–286 μ wide. Rostellum comprises a distinctly outlined somewhat hyaline, medulla (83–93 × 71–78 μ) and a less well defined, apparently muscular, cortex (104–150 × 75–98 μ); armed with 20 hooks in two rows. Front hooks 60–72 μ long, 30–36 μ from tip of handle to tip of guard, 36–41 μ from tip of guard to tip of blade; corresponding measurements of back hooks—35–42, 18–21, 21–23, μ. Neck very short.

Dorsal and ventral excretory canals approximately equal in size. Relationship of genital ducts to excretory vessels not observed. Genital pores unilateral, sinistral, pre-equatorial, and small as compared with the atria. Genital atrium in a mature segment, 50 μ deep and 38 μ in longitudinal diameter (exclusive of special pouches mentioned below); provided with 4 very prominent claw-like genital spines about 21 μ long, located 2 in each of 2 special pouches situated one in front of, and one behind, the vaginal pore.

Testes, 4 in number (no variation in number observed), spheroidal, 45–68 μ in diameter when mature, grouped in central portion of segment posterodorsal to female organs; still recognizable in gravid proglottids, arranged in a transverse row along posterior side. Cirrus pouch, 75 × 45 μ, ellipsoidal, extends mediosubanteriad to near middle of segment, contains cirrus in distal, and a loop of vas deferens in proximal, portions. Cirrus narrowly ellipsoidal, 41 × 18 μ, armed with numerous fine, porally directed, hair-like spines. Vas deferens loosely coiled anteroventrally in median portion of segment. Vasa efferentia not observed.

Mature ovary 112 μ in maximum diameter, transversely elongate, bilobate—the right and left lobes being connected by a slender elongate isthmus; dextral lobe usually further subdivided into anterior and posterior lobules. Vitellarium broadly ellipsoidal, transversely elongate, behind ovary near center of proglottid, about 37 μ in transverse diameter. Mehlis' gland situated dorsally between vitellarium and left lobe of ovary. Uterus at first a small indistinct sac; later, it becomes roughly U-

Received for publication, August 12, 1940.

* Contribution, No. 80, from the Zoological Laboratory, Oklahoma Agricultural and Mechanical College, prepared under the direction of R. Chester Hughes.

shaped, the limbs being directed backward and variously marked by constrictions. Proximal portion of vagina thick walled and dilated near the atrium, located immediately dorsal to cirrus pouch; distal end of vagina expanded as a thin-walled spheroidal seminal receptacle about 23 μ in diameter.

Host: Ardea herodias herodias Linnaeus, 1758.

Habitat: Small intestine.

Locality: Cromwell, Oklahoma.

Cotype specimens: In toto mounts of two entire strobilae and numerous fragments in U. S. Nat. Mus. Helm. Coll.

The genus *Gryporhynchus* von Nordmann, 1832, now contains 4 known species—(1) *G. pusillus* von Nordmann, 1832 (synonym, *Taenia macropeos* Wedl, 1855); (2) *G. cheilancristrotus* (Wedl, 1855) Ransom, 1909; (3) *G. macrorostratus* (Fuhrmann, 1907) Ransom, 1909; and (4) *G. tetrorchis* n. sp. *G. macrorostratus* was taken from a passeriform host, the others from CICONIIFORMES. The new species is apparently the first to be reported from the western hemisphere. Used in this study were the compilatory descriptions of Joyeux and Baer (1936) of all 3 of the old species, the original account of Fuhrmann's species, and a description by Meggitt (1931) of worms identified reservedly as "*G. macropeos* (Wedl, 1856)."

The new species differs from all the others in having fewer testes, a relatively shorter cirrus pouch, a new world distribution, and, apparently, in the U-shape of the gravid uterus. Further, *G. tetrorchis* is considerably larger than *G. pusillus*, considerably smaller than *G. cheilancristrotus*, and unlike *G. macrorostratus* in having 4 instead of 2 genital spines in the atrium. According to diagnoses of *Gryporhynchus* by Ransom (1909) and Meggitt (1924) the testes are 6–8 in number; Meggitt (1931), however, found 5 in his above-mentioned specimens of "*G. macropeos*."

BIBLIOGRAPHY

FUHRMANN, O. 1907 Bekannte und neue Arten und Genera von Vogeltänien. Centr. Bakt. I Abt. Orig. **45**: 516–536.

JOYEUX, C. AND BAER, J. G. 1936 Cestodes. Faune de France **30**: 1–613.

MEGGITT, F. J. 1924 The cestodes of mammals. London. 282 pp.

———1931 On cestodes collected in Burma. Part II. Parasitology **23**: 250–263.

RANSOM, B. H. 1909 The taenioid cestodes of North American birds. Bull. U. S. Nat. Mus. **69**: 1–141.

EXPLANATION OF PLATE, p. 174

All figures concern *Gryporhynchus tetrorchis* n. sp. and all were drawn with the aid of a camera lucida, minor details added free-handedly.

ABBREVIATIONS

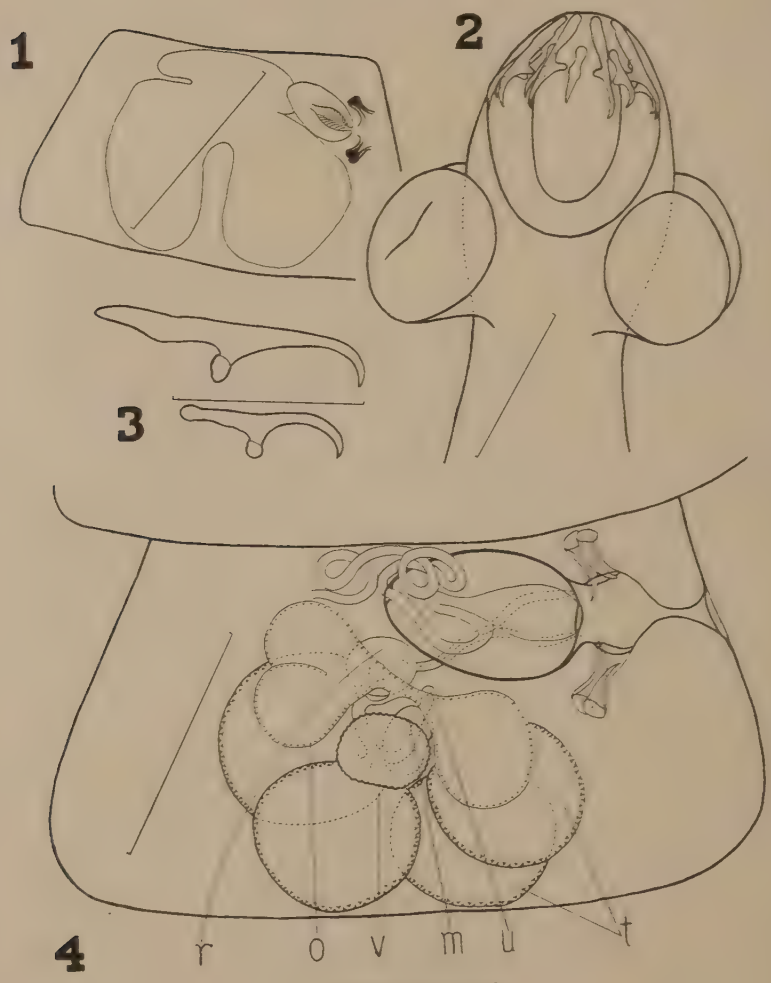
m—Mehlis' gland
o—ovary
r—seminal receptacle
t—testes
u—uterus
v—vitellarium

FIG. 1. Partially gravid proglottid. Note hair-like spines in cirrus and U-shape of uterus. Scale = 200 μ.

FIG. 2. Scolex with rostellum everted. Scale = 100 μ.

FIG. 3. Rostellar hooks, front and rear. Scale = 50 μ.

FIG. 4. Mature proglottid, ventral view, somewhat diagrammatic. Scale = 100 μ.



Gryporhynchus tetrorchis

HELMINTHS OF MUSKRATS IN SOUTHEAST TEXAS

Asa C. Chandler
The Biological Laboratory, Rice Institute, Houston, Texas

In connection with an ecological study of native fur animals in Southeast Texas conducted by Mr. Dan W. Lay, Regional Game Manager, Texas State Game, Fish and Oyster Commission, 36 muskrat carcasses were made available for parasitological examination. Sixteen of the animals were collected from Big Hill Bayou, near Port Arthur in Jefferson Co., and 20 were collected a few miles northeast of High Island in Chambers Co.

As will be seen from the accompanying table there were some striking differences in the parasite fauna of the rats from these two localities. Mr. Lay states that both localities are meadow-like marshes similar in general appearance, and have about equal muskrat populations. In the Jefferson Co. locality, however, the water is only slightly brackish, fresh enough for rice cultivation and to support a good growth of *Cabomba caroliniana,* whereas in the Chambers Co. locality the water is tidal and brackish as indicated by the presence of *Ruppia maritima.* The absence of the filarial worm, *Litomosoides carinii,* from the Chambers Co. muskrats may possibly be due to failure of the unknown intermediate host to breed in brackish water. The presence of *Nudacotyle novicia* in the brackish water area and not in the area with fresher water, however, probably cannot be accounted for in a similar manner, since this fluke has hitherto been reported from an inland locality.

The helminths of muskrats were first extensively studied by Barker (1915) in Nebraska. Since then one or a few specimens of this host have been examined in various parts of this country and in Europe, and a considerable number of new species of helminths added. The localities represented include the Great Lakes area (Barker, 1916; Price, 1931; Ameel, 1932; Olivier, 1938; Dikmans, 1935; Olsen, 1939), Maryland (Price, 1931; Smith, 1938), Great Britain (Warwick, 1936), and Russia (Schulz et al, 1933).

The parasitic fauna of the Texas muskrats is notably different from that recorded in any of these localities. Only two of the 22 species of flukes hitherto recorded from muskrats, and one new species, were found, namely: *Echinochasmus schwartzi,* hitherto recorded once, from Maryland, by Price, 1931; *Nudacotyle novicia,* hitherto recorded once, from Minnesota, by Barker, 1916; and *Phagicola lageniformis* n. sp. Of these *Echinochasmus schwartzi* was the commonest helminth, both from the standpoint of incidence and of numbers, in hosts from both localities.

Received for publication, August 19, 1940.

TABLE 1.—*Helminth parasites of muskrats in southeast Texas*

Parasites	Jefferson Co. 16 rats examined		Chambers Co. 20 rats examined	
	No.	%	No.	%
Trematodes				
Echinochasmus schwartzi	11	69	10	50
Phagicola lageniformis	0	0	1	5
Nudacotyle novicia	0	0	6	33
Nematodes				
Strongyloides ratti var. *ondatrae*	0	0	1	5
Longistriata dalrymplei ?	0	0	1	5
Rictularia ondatrae	1	6	2	10
Litomosoides carinii	4	25	0	0
Cestodes	0	0	0	0

Nudacotyle novicia occurred in 6 of 20 rats collected in Chambers Co., and in considerable numbers, but was entirely absent from the 16 rats collected in Jefferson Co. *Phagicola lageniformis* n. sp. was found in moderate numbers only in one rat, from Chambers Co.

Four species of nematodes were collected, of which three are definitely new from this host. One, *Rictularia ondatrae* n. sp., was found in three of the 36 rats examined, and was represented in both localities. *Strongyloides ratti* var. *ondatrae* n. var. was found in a single rat from Chambers Co., in small numbers. In the same rat were found two female specimens of a heligmosome which could not be positively identified in the absence of males, but which is probably *Longistriata dalrymplei* Dikmans, 1935. In 4 of the 16 rats from Jefferson Co., but in none from Chambers Co., were found adults and microfilariae of *Litomosoides carinii* (Travassos, 1919), a species common in cotton rats, *Sigmodon hispidus*, in Texas and Mexico. No tapeworms, either larvae or adults, were encountered.

Descriptions of the new species and variety follow.

Phagicola lageniformis n. sp.

(Figs. 1–2)

Very small flask-shaped flukes; posterior portion almost spherical, preacetabular portion drawn out into a slender neck about as long, or longer, than the posterior portion. Length 0.45 to 0.63 mm; maximum diameter of posterior portion 0.23 to 0.26 mm; diameter of neck at narrowest point only 45 to 65 μ. Dorsal lip well developed, rounded. Oral sucker 45 to 55 μ in diameter, surrounded by a crown of 18 spines, 16 of them in a single circle, two situated more posteriorly on dorsal side; spines 18 to 20 μ long, all approximately equal in size. Oral diverticulum broad at base, tapering to a narrow and usually twisted end; length 90 to 140 μ, in most specimens reaching only from half to two-thirds distance from oral sucker to pharynx. Pharynx 30 to 33 μ wide and 33 to 38 μ long, its anterior rim situated 120 to 220 μ from anterior end, depending upon degree of contraction; esophagus very short; ceca wide, not traceable behind level of acetabulum, where they probably end. Acetabulum 40 to 50 μ in diameter, not obviously sunk in genital sinus, situated at or slightly behind middle of body length.

Testes situated at or near posterior end of body, usually transversely elongated, varying from 60 by 70 μ to 35 by 90 μ. Ovary slightly smaller than testes, situated in front of right testis. Seminal receptacle very large, median, in front of testes and at left of ovary. Vitellaria consist of two groups of usually 7 follicles each, situated at

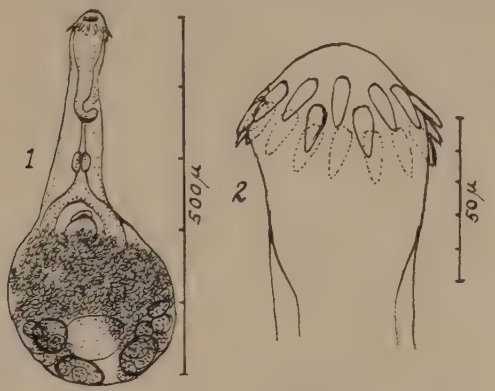


Fig. 1. *Phagicola lageniformis* n. sp. Ventral view.
Fig. 2. *Phagicola lageniformis* n. sp. Cephalic crown of spines.

sides of posterior part of body, partly overlying the testes and extending forward to the level of the ovary. All available space between posteriorly situated reproductive organs and acetabulum occupied by irregular coils of the uterus filled with eggs. Eggs 20 to 21 by 10 to 12 μ.

Host: Muskrat.
Habitat: Intestine.
Locality: Southeast Texas.
Type specimen: Deposited U. S. N. M., Helm. Coll.

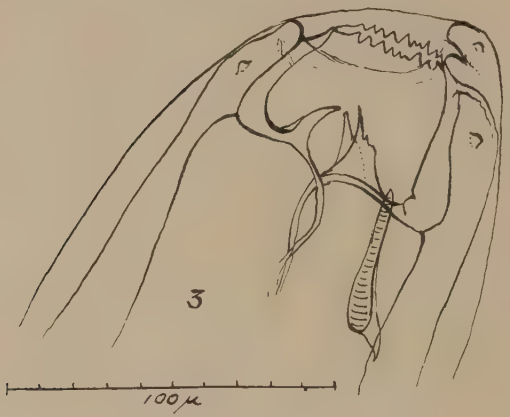
This species resembles *P. nana* (Ransom, 1920) and *P. angrensis* (Travassos, 1916) in the number and arrangement of the oral spines, but differs from *nana* in shape of body, size of oral diverticulum, and size of spines, and from *angrensis* in shape of body and in length of the prepharyngeal region, which in *angrensis* is very short, resulting in the oral diverticulum reaching beyond the pharnyx in that species.

Rictularia ondatrae n. sp.

(Figs. 3–4)

Females large coarse worms, males relatively very small. Mouth terminal, but with a liplike projection of the ventral wall; buccal capsule heavily chitinized with three sharp teeth, jagged in some specimens, at the bottom, and a ring of about 18 denticles on its rim. Two pairs of papillae on the ventral side, one pair on dorsal side. Anterior end of first comb at level of bottom of capsule.

Females up to 28 or 30 mm long, with a maximum diameter, behind middle of body, of 850 to 900 μ. Mouth cavity about 60 μ across at anterior end, widening out to about 90 μ at bottom; about 85 μ deep. Diameter of head where cuticle bulges behind mouth capsule about 240 μ. Combs 24 or 25 to end of esophagus, 32 to vulva; total number of combs and spines 73 to 75. Combs in esophageal region contiguous, the backward-projecting spines very small, the pointed anterior ends of the bases much more conspicuous; cuticular processes in vulvar region transitional between combs and spines; projecting part of largest spines, about 1 mm behind vulva, 50 μ long, from here gradually decreasing in size and becoming spaced farther and farther apart, the last spines extremely small, over 1 mm apart, and ending about 2.5 to 3 mm from end of tail. Esophagus 3.2 to 3.5 mm long. Vulva 0.5 to 1 mm behind end of



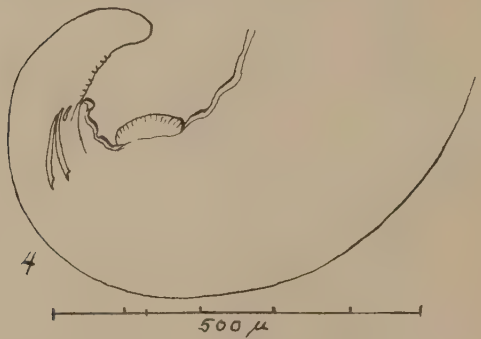


FIG. 3. *Rictularia ondatrae* n. sp. Head of male.
FIG. 4. *Rictularia ondatrae* n. sp. Caudal end of male.

esophagus. Vagina turns posteriad, and bifurcates about 300 to 400 μ behind vulva. Tail conical, rounded at tip, 500 to 630 μ long. Eggs 50 to 52 by 37 to 38 μ.

Males 6 to 7 mm long, with maximum diameter of about 420 μ. Esophagus 1.5 mm long. Mouth cavity 32 μ across at opening, 50 μ across at bottom, 40 μ deep. Combs contiguous for about one-third of body length, then becoming more and more widely spaced, but only about 200 μ apart, point to point, even at posterior end of body, and not decreasing in size very much. Last spines about 350 μ anterior to anus. Total number of combs and spines 52. Anus about 150 μ from tip of tail. Spicules nearly equal, one 110 μ long, the other about 98 μ; gubernaculum 20 μ long. A large median papilla just anterior to cloaca; no preanal papillae seen; four pairs of postanal papillae about equally spaced, last one about midway between cloaca and tip of tail. One fanlike expansion of the cuticle on midventral line, beginning 90 to 100 μ anterior to cloaca, 90 μ long and 30 μ broad.

Host: Muskrat.
Habitat: Intestine.
Locality: Southeast Texas.
Type specimen: Deposited U. S. N. M., Helm. Coll.

As noted by Hall (1913) the species of *Rictularia* fall into two well-

TABLE 2.—*The following table shows the distinctions between species of* Rictularia *so far described from North America*

	splendida Hall, 1913	*coloradensis* Hall, 1916	*scalopis* Goodrich, 1932*	*halli* Sandground, 1935	*onychomis* Cuckler, 1939	*ondatrae* Chandler, 1940
Length	♀ 8–10.5 mm ♂ 4.8 mm	♀ 9–10 mm ♂ 3 mm	♀ 54 mm ♂ unknown	♀ 47 –70 mm ♂ 3.7– 7.8 mm	♀ 24–42 mm ♂ Unknown	♀ 25–30 mm ♂ 7 mm
Total, combs and spines, ♂	108–109	42	Unknown	37–38	Unknown	52
Same, ♀, prevulvar and postvulvar	55 + 83	31 + 33 +	28 + ?	25–28 + 5–10	33 + 23–27	32 + 41–43
Position of vulva relative to end of esophagus	Just behind	Opposite	2 mm behind	Just behind	Anterior	0.5 to 1 mm behind
Position of first comb relative to buccal capsule	Own length behind	16 μ behind	93 μ behind	Just behind	100 μ behind	Reach to it
Length of spicules	207 μ, 207 μ	145 μ, 180 μ	Unknown	40–50 μ, 95–100 μ	Unknown	98 μ, 110 μ
Ventral fans (♂)	8	None	Unknown	4 or 5	Unknown	One
Host	Coyote	Chipmunk	Mole	Chipmunk	Grasshopper mouse	Muskrat

* *R. citelli* McLeod, 1933, from two species of *Citellus* in Manitoba is not sufficiently well described to distinguish it from *R. scalopis* Goodrich, 1932. The females of *R. citelli* are 30–60 mm long, with 28 prevulvar spines; a single male was 14 mm long, had equal spicules 117 μ long, and no ventral fans.

defined groups: (1) those from carnivores, the females of which have over 100 pairs of combs and spines, of which the majority are combs, and in which there is no well-defined transition from combs to spines in the region of the vulva, and (2) those from rodents, insectivores and bats, in the females of which there are less than 100 combs and spines, and a fairly sharp transition from one to the other in the region of the vulva. In many species the male is unknown, and this is true of two of the five previously known forms from North America (*R. scalopis* and *R. onychomis*). Fortunately there are good characters for separating the females, and Cuckler (1939) devised a key based on this sex alone, though he omitted *R. halli* Sandground, 1935. *R. ondatrae* differs from all the other North American forms of the rodent-insectivore-bat group in having a total of over 70 combs and spines, and 32 prevulvar combs. It differs widely from the forms in which males are known in the size of the spicules, and in the number of ventral fans anterior to the cloaca. Other differential characters are shown in the accompanying table. The present species comes nearest to *R. scalopis* Goodrich, 1932, but is smaller, has a much less prominent ventral lip in front of the oral capsule, has the first comb more anterior in position, has the vulva nearer to the end of the esophagus, and has a larger number of prevulvar combs. The total number of combs and spines in *R. scalopis* is not given, and the male is unknown.

Strongyloides ratti var. *ondatrae* n. var.

Only a few specimens of this parasite were found. It resembles *S. ratti* in most respects but is longer and much more slender than the typical form as described by Sandground (1925). The variety from the muskrat is 4 mm long with a diameter of only 33 μ, 1/120 of the length, whereas the size of typical *S. ratti* as given by Sandground is 1.85 to 3.03 mm with a diameter of 40 μ, 1/60 of the length. In *ondatrae* the anus is 55 to 60 μ from the posterior end, and the vulva divides the body 2.5 : 1.5. The esophagus is 1 mm long, and the eggs measure 45 by 25 μ.

Host: Muskrat.
Habitat: Intestine.
Locality: Southeast Texas.

SUMMARY

Thirty-six muskrats collected in southeast Texas yielded 3 species of trematodes, four of nematodes, and no cestodes. One trematode, *Phagicola lageniformis,* is new; of the nematodes, *Rictularia ondatrae* is new; *Strongyloides ratti* var. *ondatrae* is a new variety; and *Litomosoides carinii* is a new host record.

BIBLIOGRAPHY

AMEEL, D. J. 1932 The muskrat, a new host for Paragonimus. Science, **75**: 382.
BARKER, F. D. 1915 Parasites of the American muskrat (*Fiber zibethicus*). J. Parasitol. **1**: 184–197.
———1916 A new monostome trematode parasitic in the muskrat, with a key to the parasites of the American muskrat. Tr. Am. Micr. Soc. **35**: 175–184.

CUCKLER, A. C. 1939 *Rictularia onychomis* n. sp. (Nematoda, Thelaziidae) from the grasshopper mouse, *Onychomys leucogaster* (Weid). J. Parasitol. **25**: 431–435.

DIKMANS, G. 1935 New nematodes of the genus Longistriata in rodents. J. Wash. Acad. Sc. **25**: 72–81.

GOODRICH, A. L. 1932. *Rictularia scalopis,* sp. nov., a nematode from the mole, *Scalops aquaticus* (Linn.). Tr. Am. Micr. Soc. **51**: 216–218.

HALL, M. C. 1913 A new nematode, *Rictularia splendida,* from the coyote with notes on other coyote parasites. Proc. U. S. Nat. Mus. **64**: 73–84.

————1916 Nematode parasites of mammals of the Orders Rodentia, Lagomorpha, and Hyracoidea. Proc. U. S. Nat. Mus. **50**: 1–258.

OLIVIER, L. 1938 A new trematode, *Allassogonoporus marginalis,* from the muskrat. J. Parasitol. **24**: 155–160.

OLSEN, O. W. 1939. Schizotaeniasis in muskrats. J. Parasitol. **25**: 279.

PRICE, E. W. 1931 Four new species of trematode worms from the muskrat, *Ondatra zibethica,* with a key to the trematode parasites of the muskrat. Proc. U. S. Nat. Mus. **79**(Art. 4): 1–13.

RANSOM, B. H. 1920 Synopsis of the trematode family Heterophyidae, with descriptions of a new genus and five new species. Proc. U. S. Nat. Mus. **57**: 527–573.

SANDGROUND, J. H. 1925 Speciation and specificity in the nematode genus Strongyloides. J. Parasitol. **12**: 59–80.

————1935 *Spirura michiganensis* n. sp. and *Rictularia halli* n. sp., two new parasitic nematodes from *Eutamias striatus lysteri* (Richardson). Tr. Am. Micr. Soc. **54**: 155–166.

SCHULZ, R. E., ORLOW, J. W. AND KUTASS, A. J. 1933 Zur Systematik der Subfamilie Synthetocaulinae Skrj., 1932, nebst Beschreibung einiger neuer Gattungen und Arten. Zool. Anz. **102**: 303–310.

SMITH, F. R. 1938 Muskrat investigations in Dorchester Co., Maryland, 1930–1934. U. S. Dept. Agric., Circ. **474**: 1–24.

TRAVASSOS, L. 1930 Revisão do Genero Ascocotyle Looss, 1899. Mem. Inst. Oswaldo Cruz **23**: 61–97.

WARWICK, T. 1936 Parasites of the muskrat (*Ondatra zibethica* L.) in the British Isles. Parasitology **28**: 395–401.

RESEARCH NOTES

TWO NEW TREMATODES FROM THE BONITO, *SARDA SARDA*, IN THE GULF OF MEXICO

A specimen of bonito, *Sarda sarda*, taken off the Texas coast near Freeport, yielded two new species of trematodes, one a gasterostome of the genus *Rhipidocotyle*, the other a hemiurid of the genus *Sterrhurus*. Descriptions of them follow:

Rhipidocotyle angusticolle n. sp.

(Figs. 1–3)

Body elongate, broadest at about level of ovary; anterior portion constricted to a narrow neck just behind sucker. Anterior end of very peculiar shape with two hornlike projections on each side dorsally, a pair of lobes overhanging the sucker ventrally, and the sucker itself projecting ventrally and posteriorly like a chin. Length 1.2 to 1.4 mm; maximum diameter 300 to 420 μ; diameter of neck about 65 μ, its depth only about 30 μ; diameter across dorsal horns about 170 μ; length of sucker about 110 μ. Pharynx about 60 μ in diameter; intestinal sac about 190 μ long and 135 μ wide, situated behind middle of body. Ovary at level of posterior portion of intestinal sac, at right, about 65 to 70 μ in diameter; testes one behind the other, posterior to the ovary, about 120 to 130 μ in diameter. Cirrus sac about 400 μ long; copulatory bursa about 125 μ in diameter. Vitellaria in two clusters of about 15 follicles each, the follicles about 40 μ in diameter, situated at level of anterior part of intestinal sac and anterior to it; vitelline ducts unite at level of junction of two testes. Uterus occupies available space behind intestinal sac and to left of testes; eggs about 21 to 22 μ long and 14 to 16 μ broad.

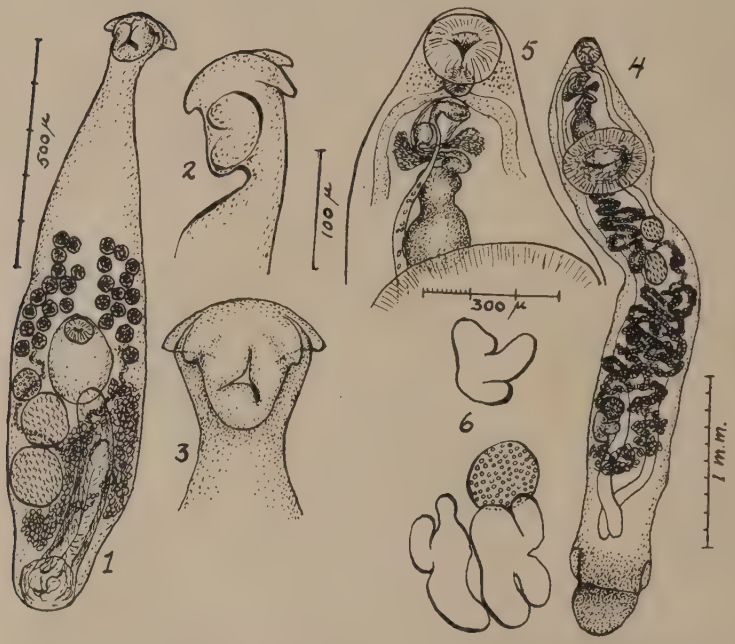
This species differs from all of the nine members of the genus hitherto described from marine fishes (see Eckmann, 1932; Chandler, 1935; Manter, 1940) by the very slender neck, deeply indented ventrally behind the anterior sucker, the posterior edge of which projects chinlike, and in the peculiar shape of the cephalic hood, with its prominent horns projecting dorso-laterally

Sterrhurus texanus n. sp.

(Figs. 4–6)

Body elongate, cylindrical for most of its length, but bulging markedly at level of ventral sucker, and tapering anterior to it; retractile appendage only about 1/7 of body length. Length 3.25 to 3.6 mm; breadth behind ventral sucker about 0.5 mm, depth about 0.4 mm. Retractile appendage rounded, about 0.5 mm long. Oral sucker 150 to 180 μ in diameter; ventral sucker very large, over three times as broad as oral sucker measuring 550 μ in breadth, 400 to 500 μ antero-posteriorly, and about 380 μ dorso-ventrally. Distance between suckers 350 μ or less. Pharynx about 70 μ in diameter; no prepharynx. Intestinal ceca extend almost to retractile appendage. Genital opening nearly median, close behind oral sucker. Sinus sac about 150 μ long and 110 μ broad; at its inner end a spherical prostatic vesicle, just in front of which the metraterm enters to form a distended hermaphroditic duct. Outside sinus sac vas deferens at first narrow, with prostate glands spreading winglike on either side of it, then forming a large trilobed seminal vesicle, 280 μ long with a maximum breadth of 145 μ. Testes round or oval, situated slightly oblique, about 2/5 body length from anterior end, separated by a loop of the uterus; size from 100 × 130 to 130 × 200 μ. Ovary and yolk glands closely associated, widely separated from testes; ovary about 150 μ in diameter; vitelline glands, each with three or four lobes, situated on right side of body. Uterus in more or less horizontal loops behind ventral sucker, the loops extending some distance behind the vitelline glands; eggs about 18 by 11 to 12 μ.

A considerable number of species of *Sterrhurus* have been described from marine fishes by Looss, Linton, Nicoll, Manter, Stunkard and Nigrelli, Park, and



FIGS. 1 to 3: *Rhipidocotyle angusticolle* n. sp. 1, ventral view; 2, lateral view of anterior end; 3, ventral view of anterior end.

FIGS. 4 to 6: *Sterrhurus texanus* n. sp. 4, ventral view; 5, anterior end; 6, vitelline glands: upper, one gland in ventral view; lower, complex of two glands and ovary seen from right side.

Yamaguti, and there is considerable doubt as to whether all of them are valid species. Looss's (1907) type species, *musculus,* Manter's (1934) *floridensis,* and the forms assigned by Linton in 1910 to his *monticellii* (1898) are quite possibly identical, as Stunkard and Nigrelli (1934) pointed out. The species here described is a larger and much more slender form than these, with the ventral sucker much larger relative to the width, with a very short "tail," and with the ovary and vitelline gland complex situated far behind the testes, which are almost tandem in position and at some distance behind the ventral sucker. These characters, together with the blunt lobing of the vitellaria, the large tripartite seminal vesicle, and the characters of the sinus sac, distinguish this form from all members of the genus except *imocavus,* described by Looss (1907) from *Thynnus* in Egypt, which it closely resembles. It is, however, larger than *imocavus,* with less prominent dorsal lip, larger vitellaria relative to the ovary, and larger eggs.—ASA C. CHANDLER, *Rice Institute, Houston, Texas.*

A NEW SPIRUROID NEMATODE, *HABRONEMA AMERICANUM,* FROM THE BROAD-WINGED HAWK, *BUTEO PLATYPTERUS*

A few specimens of a *Habronema* which proved to be a new species were collected from the stomach of *Buteo platypterus* in Cherokee Co., Texas, by Mr. Rollin H. Baker, Field Biologist with the Texas Game, Fish and Oyster Commission. The specific description follows:

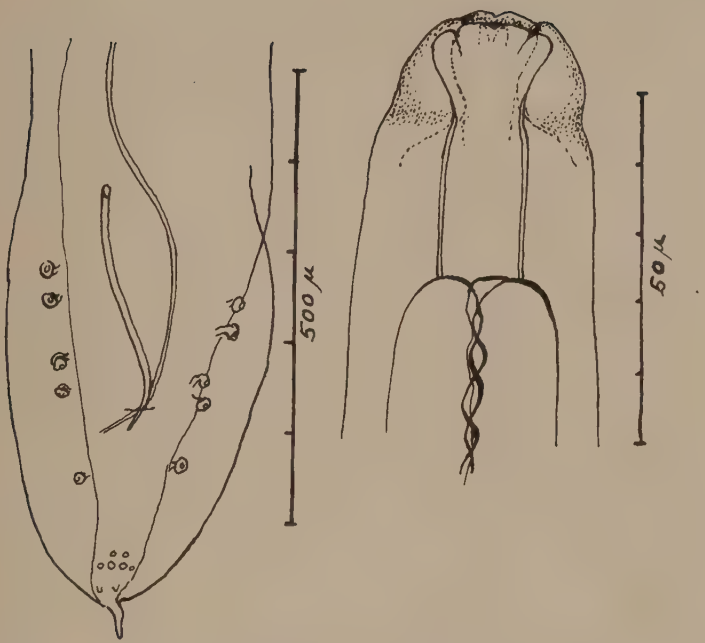


FIG. 1. *Habronema americanum* n. sp. Left, tail of male; right, head of female.

Habronema americanum n. sp.

Body elongate, strongly striated, with lateral alae, one better developed than the other, extending greater part of length of body. Cervical papillae anterior to nerve ring. Head indistinctly set off from body, about 35 μ in diameter. Lateral lips broad, not very distinctly lobed; dorsal and ventral lips bluntly pointed. Bottom of oral vestibule about 38 to 40 μ from anterior end, diameter about 13 to 15 μ. Esophagus about 3 to 3.5 mm long.

Male about 10 mm long and 260 μ in diameter. Caudal alae unequal, the right one longer, marked by wavy longitudinal striations. Four pairs of pedunculated preanal papillae, one similar pair postanal, and 8 very minute sessile papillae near tip of tail, arranged 2, 4, 2. Tail terminated by a fleshy spike about 30 μ long. Spicules unequal, the right one 225 to 250 μ long, the left one 725 to 750 μ long; gubernaculum 26 μ long.

Female about 15 mm long and 350 μ in diameter. Tail conical, 325 μ long. Vulva anterior to middle of body, dividing it about 2 : 3. Eggs elliptical, 37 to 40 μ long and 18 to 19 μ broad.

This species closely resembles *H. leptoptera* (Rudolphi, 1819) of Old World hawks, differing principally in the shape of the lips. Other minor differences are in the greater length of the esophagus, the longer spicules, and the different arrangement of the sessile papillae at the end of the tail of the male.—ASA C. CHANDLER, *Rice Institute, Houston, Texas.*

THE OCCURRENCE OF *HEXAMITA* (*OCTOMITUS*) *COLUMBAE* IN PIGEONS IN CALIFORNIA

Nöller and Buttgereit (Centr. Bakt. I Abt. Orig. **75**: 239–240) and Buttgereit (Inaug. Dissert., Tierärstl. Hochs., Berlin) in 1923 reported a catarrhal enteritis of

young pigeons in Germany. The enteritis extended throughout the intestine and was accompanied by large numbers of a small flagellate (5–9 μ by 2.5–3.5 μ) of the genus *Hexamita* (*Octomitus*). The organisms were increasingly abundant in the lower jejunum and ileum, and most abundant 5 cm anterior to the vent. They were abundant not only in the lumen, but also deep in the crypts of the villi. There was marked hyperemia of the duodenum and jejunum, and an infiltration of lymphocytes. They named this species *Hexamita* (*Octomitus*) *columbae*.

It is the purpose of the present paper to describe four similar cases and to report *Hexamita columbae* from California. To our knowledge it is the first time this condition has been reported from pigeons in the United States.

Case I.—The first time that *Hexamita* was observed by us in pigeons was in a bird brought to the State Department of Agriculture Poultry Pathological Laboratory at Los Angeles. This case is referred to by Niemeyer (1939, J. Am. Vet. Med. Assn. **94**: 434–435), and came from a flock which was also suffering from paratyphoid infection and trichomoniasis of the crop. The relationship of *Hexamita* to the mortality in this flock was not determined. The intestine of the one bird examined showed no marked changes. No *Hexamita* were found in the duodenum, a few were present in the jejunum and ileum, large numbers were found in the rectum and a few in the bursa of Fabricius. The absence in the upper intestine and the lack of pathology suggests that this bird was in the carrier stage rather than suffering from the presence of this parasite.

Case II.—The second case was a group of five birds from a flock of tumbler pigeons. Several of the young birds and one breeder in this flock were in poor condition and thought by the owner to be suffering from coccidiosis. Oöcysts were found in only one bird and then in very small numbers.

Hexamita was found in four of the five birds; the fifth was suffering from impaction of the rectum. Of the four birds, two were suffering from a catarrhal enteritis. The duodenum was fairly normal, but the jejunum was hyperemic and the wall of the ileum was thin. There were several diphtheritic ulcers in the rectum. *Hexamita* was absent or rare in the duodenum, numerous in the jejunum and ileum and most abundant in the rectum. It was present also in the bursa of Fabricius. No pathogenic bacteria could be isolated from these birds.

Case III.—The third case was brought in by the owner of the birds mentioned above. It was a banded bird which had escaped from some other loft and had been present in his loft for only a day or two. This bird was in poor flesh. There was marked catarrhal enteritis throughout the intestine and ulceration of the ileum and rectum. *Hexamita* was abundant in scrapings from the jejunum, ileum, and rectum. All of these cases originated in Southern California.

Case IV.—The fourth outbreak occurred in a loft located in the San Francisco Bay region. The owner reported heavy losses in his young squabs with occasional losses in the adults. A total of five pigeons of various ages were examined. All were in fair flesh, and three were suffering from a marked case of trichomoniasis of the upper digestive tract. Coccidia and a few ascarids as well as *Hexamita* were found in the intestines. The *Hexamita* were present in large numbers in the posterior two-thirds of the intestines and in small numbers in the bursae of Fabricius. In one young squab a small rectal ulcer was present and *Hexamita* in large numbers were clinging to the tissues scraped from this lesion. Because of the numbers of other parasites also present in these birds it was difficult to determine which species were responsible for the losses.

That *Hexamita columbae* is not merely a ubiquitous commensal in pigeons is evidenced by the fact that healthy pigeons have been examined from four other flocks and *Hexamita* was not present.

The disease described by Nöller and Buttgereit in Germany and also encountered in these birds in California is similar to infectious catarrhal enteritis of turkey poults (Hinshaw, McNeil, and Kofoid, 1938, Cornell Vet. **28**: 281–293; McNeil, Hinshaw, and Kofoid, 1941, Am. J. Hyg. in press). One essential difference in the pathology of hexamitiasis of pigeons is the consistent catarrhal inflammation with a heavy mucus deposit in the rectum of young pigeons. This was also noted by Butt-

gereit (1923). The owner of the loft from which the pigeons of Case II came, reports that he has had only a small mortality in his young birds, but many of them are unthrifty. No other infection has been found on this ranch; so it is highly probable that *Hexamita columbae* is responsible for the loss he has had and for the stunting of the squabs.

It has not been possible to infect turkey poults with *Hexamita columbae* (cf. McNeil, Hinshaw, and Kofoid, l.c.), nor is it likely that it is possible to infect pigeons with *Hexamita meleagridis,* since the pigeons examined by us and found free from *Hexamita* were on ranches where *Hexamita meleagridis* was abundant in turkeys, and where they frequented the infected pens.—E. McNeil and W. R. Hinshaw, *University of California, Division of Veterinary Science, Davis, California.*

CERCARIA ELONGATA BRACKETT, 1940, FROM A NEW SNAIL HOST, *MENETUS EXACUOUS* (SAY), IN MINNESOTA

A schistosome cercaria identified as *Cercaria elongata* Brackett, 1940 (J. Parasitol. **26**: 195–200), was found in 2 of 40 specimens of the planorbid snail, *Menetus exacuous* (Say), collected June 22, 1940, from Lake Francis, Isanti Co., Minnesota. A small number of *Gyraulus parvus* (Say), the host Brackett reports, collected on the same date and on Sept. 9, 1940, from this lake were also positive for *C. elongata.* Although Brackett was unable to secure more than one dermatitis lesion experimentally, numerous characteristic dermatitis lesions were obtained by the writers. On one occasion when a vial containing 15 *C. elongata* from *M. exacuous* was placed in contact with the skin of the inner surface of the forearm for 12 minutes all of the cercariae penetrated and typical schistosome dermatitis lesions developed. Certainly, as Brackett concludes, this cercaria and its hosts will have to be considered in future studies of outbreaks of "swimmer's itch".—Ashton C. Cuckler and Lawrence R. Penner, *Department of Zoology, University of Minnesota, Minneapolis.*

JOHN EARL GUBERLET
(1887–1940)

IN MEMORIAM

John Earl Guberlet (1887–1940)

John Earl Guberlet[1] was born at Courtland, Nebraska, March 18, 1887, the son of a farmer. He attended Bethany College at Lindsborg, Kansas, (A.B., 1909) where he studied zoology under Prof. Emil O. Deere, though for a time he was undecided as between medicine, zoology, or the ministry. The year following his graduation he spent at the University of Colorado. Here Prof. T. D. A. Cockerell interested him in the parasitology which eventually became his life-work. In 1910 Guberlet went to the University of Illinois. He worked first for his A.M. under Prof. J. S. Kingsley, his thesis being "On the Osteology of the Loricati" or rock fishes. Then he turned to parasitology for his doctorate under Prof. H. B. Ward, investigating the cestodes of poultry. He received his Ph.D. in 1914.

The year 1914–15 Guberlet was instructor in zoology at the University of Oregon, and from 1915 to 1918 professor of biology at Carroll College, Waukesha, Wisconsin. In 1918 he went to the Oklahoma Agricultural and Mechanical College at Stillwater, Oklahoma, as assistant parasitologist in the agricultural experiment station and assistant professor of zoology, being promoted in 1921 to parasitologist and full professor. In Oklahoma he went on with his parasitological investigations, specializing on the worm parasites of domestic animals.

Guberlet was called to the University of Washington at Seattle in 1923, where he was advanced to a full professorship in 1930. Beginning in 1924, he spent his summers at Friday Harbor, first on the staff of the Puget Sound Biological Station and then, after 1930, on the staff of the Oceanographic Laboratories of the University.

During these years in Seattle and at Friday Harbor, Guberlet's researches followed two main lines. He became associated with the course in fish diseases offered by the University's School of Fisheries. This led to studies on the parasites and diseases of fresh-water fishes, especially those in fish-hatcheries, and his investigations were of value in the development of pisciculture in the western portions of the state.

In connection with the marine work at Friday Harbor, he and his students[2] produced a notable series of studies on the flatworm parasites

[1] Or "Gutberlet," as he spelled it previous to 1919.

[2] In addition to the papers cited in the bibliography of which Guberlet was author or co-author, the following were published independently by his students:

Folda, Florence. 1928 *Megalocotyle marginata*, a new genus of ectoparasitic trematodes from the rock fish. Publications Puget Sound Biol. Sta. 6: 195–206.

Hart, John Francis. 1936 Cestoda from fishes of Puget Sound. II. Tetrarhynchoidea. Tr. Am. Micr. Soc. 55: 369–387.

of the marine fishes. One or two further papers on this subject were nearly ready for publication at the time of his death. He had built up a valuable parasitological collection and reprint library.

In the second place, Guberlet found the Puget Sound marine fauna new and strange. He was required to familiarize himself with it by virtue of the course in invertebrate embryology and zoology that he was teaching, and I have heard him tell how he was liberal with the midnight oil during his early sessions at the Biological Station. The result was that he came to have an extraordinarily extensive knowledge of the larger marine species. One result of this study was the book, "Animals of the Seashore," which he helped his wife, Muriel Lewin Guberlet, prepare for publication in 1936. This text, an illustrated account of nearly 200 of the common invertebrate animals of the Puget Sound region, while primarily intended for amateur and high school use, has proven to be of considerable value in the instructional work at the Oceanographic Laboratories. Another product of Guberlet's marine work was an extensive study of the distribution of the larger marine animals of Puget Sound, derived from many years of dredging mostly from the University's research M. S. *Catalyst*. Guberlet was at work on the final manuscript of this paper at the time of his death.

In addition to his instructional work in parasitology and marine zoology, Guberlet had charge of the pre-medical zoology course, and many generations of "pre-medics" have profited from his kindly sympathetic guidance. With even the largest classes he had the capacity for personalizing his relationship with his students. Those aspects of his personality which caused him at one time to consider the profession of the ministry came to the fore and made Guberlet's zoology one of the unforgettable experiences of the pre-medical student. The place he has left will be hard to fill.

As a colleague on the faculty, Guberlet was kindly and cooperative, active in the work of the faculty committees to which he belonged. He was a member of ΦΣΚ, ΓΑ, ΣΞ, ΦΚΦ, and ΦΣ, A.A.A.S. (fellow), American Society of Zoologists, American Microscopical Society (president, 1935), American Society of Parasitologists, American Society of Tropical Medicine, Society for Experimental Biology and Medicine, Western Society of Naturalists, and the Pacific Northwest Bird and Mammal Society (treasurer, 1930–).

During the spring of 1926 Guberlet held a visiting professorship in zoology at the University of Hawaii at Honolulu. In 1928–29 he was a delegate to the International Congress of Tropical Medicine and

———1936 Cestoda from fishes of Puget Sound. III. Phyllobothrioidea. Ibid **55**: 488–496.

Lloyd, Lowell Clyde. 1938 Some digenetic trematodes from Puget Sound fish. J. Parasitol. **24**: 103–133.

Hygiene at Cairo, Egypt. At the same time he visited many of the laboratories of western and southern Europe, especially the Stazione Zoologica at Naples and the London School of Hygiene and Tropical Medicine. In 1935 he was a delegate to the 7th American Scientific Congress in Mexico City.

Early in 1937 Dr. Guberlet was stricken by a severe attack of coronary thrombosis. By June, however, he had recovered sufficiently to resume his university duties, which he continued with hardly a day's interruption for three and one-half years. Recovery, however, had only been partial. A second attack at his Seattle home shortly after five on the morning of December 30, 1940, proved fatal within a few minutes, and he died, probably without waking.

Dr. Guberlet is survived by his wife, Muriel Lewin Guberlet, whom he married on December 29, 1915, at Lindsborg, Kansas, by a son, Frank Lewin Guberlet, and by a daughter, Florence Evangeline Guberlet.—MELVILLE H. HATCH, *University of Washington, Seattle.*

THE SCIENTIFIC PUBLICATIONS OF JOHN EARL GUBERLET[3]

1915 On the osteology of some of the Loricati. Illinois Biol. Monogr. **2**: 170–210 (5 plates, 13 Figs.).
1916 Morphology of adult and larval cestodes from poultry. Tr. Am. Micr. Soc. **35**: 23–44 (30 Figs.).
——— Studies on the transmission and prevention of cestode infection in chickens. J. Am. Vet. Med. Assn. **49**: 218–237.
1919 On the life history of the lungworm, *Dictyocaulus filaria*, in sheep. Ibid **55**: 621–627.
——— On the life history of the chicken cestode, *Hymenolepis carioca* (Magalhaes). J. Parasitol. **6**: 35–39 (4 Figs.).
1920 A new bladder fluke from the frog. Tr. Am. Micr. Soc. **39**: 142–148 (7 Figs.).
1921 Studies on the sheep stomach worm—*Haemonchus contortus* Rud. J. Am. Vet. Med. Assn. **39**: 716–721.
——— Some internal parasites of Oklahoma livestock. Oklahoma Agric. & Mech. Coll. Ext. Serv. Circ. No. 127: 3–15.
——— Stomach worms in sheep. Oklahoma Agric. Exper. Sta. Bull. No. 137: 3–16 (5 Figs.).
1922 Stomach worms in sheep. Am. Sheep Breeder and Wool Grower **42**: 265–270.
——— Some facts concerning human parasites in Oklahoma. Oklahoma State Med. J. **15**: 187–193.
——— Three new species of Holostomidae. J. Parasitol. **9**: 6–14 (13 Figs.).
——— Some notes on the parasite fauna of Oklahoma. Proc. Oklahoma Acad. Sc. **2**: 36–41 (Univ. Oklahoma Bull. N. S. 247; Univ. Stud. 15).
——— Lowery Laymon Lewis. Science **56**: 563–564.
——— Potassium nitrate poisoning in chickens. J. Am. Vet. Med. Assn. **62**: 362–365.
1923 *Hemistomum confusum*, a homonym. Tr. Am. Micr. Soc. **42**: 68.
——— An epizootic of aspergillosis in chickens. J. Am. Vet. Med. Assn. **63**: 612–620.
——— Roup in poultry. Oklahoma Agric. & Mech. Coll. Agric. Exper. Sta. Circ. July 1923. 2 pp.

[3] The first three titles were published under the name of "Gutberlet."

——— Parasites of dogs and cats of Oklahoma. Proc. Oklahoma Acad. Sc. **3**: 71–78 (Univ. Oklahoma Bull. N. S. 271; Univ. Stud. 16).
1924 Notes on the life history of *Ascaridia perspicillum* (Rud.). Tr. Am. Micr. Soc. **43**: 152–156.
1925 *Malacobdella grossa* from the Pacific coast of North America. Publications Puget Sound Biol. Sta. **5**: 1–13 (8 Figs.).
1926 Ecto-parasitic infusoria attacking fish of the Northwest. Univ. Washington Publications Fish. **2**: 1–16 (5 Figs.).
——— Notes on the parasitic fauna of Hawaii. Proc. Hawaiian Acad. Sc., First Ann. Meeting. Bernice P. Bishop Mus. Spec. Publ. **11**: 29–30.
1927 Some relationships of the parasitic flatworms of the birds of the Northwest. Murrelet **8**: 1–3 (mimeographed).
——— (with Harry A. Hansen and Jean A. Kavanagh) Studies on the control of *Gyrodactylus*. Univ. Washington Publications Fish. **2**: 17–29.
1928 Parasitic worms of Hawaiian chickens with a description of a new trematode. Tr. Am. Micr. Soc. **47**: 444–453 (5 Figs.).
——— Notes on a species of *Argulus* from goldfish. Univ. Washington Publications Fish. **2**: 31–41 (7 Figs.).
——— Observations on the spawning habits of *Melibe leonina* (Gould). Publications Puget Sound Biol. Sta. **6**: 263–270.
——— Two new genera of trematodes from a red-bellied water snake. J. Helm. **6**: 205–218 (13 Figs.).
1930 Notes on relationships of parasitic flatworms to birds and mammals. Murrelet **11**: 15–17.
1931 (with V. S. Samson and W. H. Brown.) A report of hydrocoele embryonalis, or yolk-sac disease, in salmon fry, with a note on its cause. Tr. Am. Micr. Soc. **50**: 164–167.
1932 Parasitism in Northwestern United States of North America in relationship to public health. Compt. Rend. Cong. Internat. Méd. Trop. et Hyg. (Le Caire, Déc. 1928) **4**: 41–47.
——— (with Lowell C. Lloyd) A new genus and species of Monorchidae. J. Parasitol. **18**: 232–239 (5 Figs.).
——— Notes on some Onchocotylinae from Naples with a description of a new species. Pubblicazioni Stazione Zool. Napoli **12**: 323–336 (10 Figs.).
1933 (with R. C. Miller) Notes on birds observed at sea off the North Pacific coast. Murrelet **14**: 7–8 (for correction see Ibid **14**: 46).
1934 Recent advances in our knowledge of the parasites of the marine fishes of the Pacific. Proc. 5 Pacific Sc. Cong. (Canada 1933), pp. 4165–4169.
——— Observations on the spawning and development of some Pacific annelids. Ibid, pp. 4213–4220.
1936 (with Lowell C. Lloyd) *Syncoelium filiferum* (Sars) from the Pacific salmon. Tr. Am. Micr. Soc. **55**: 44–48 (5 Figs.).
——— (with John F. Hart) Cestoda from fishes of Puget Sound. I. Spathebothriidea, a new superfamily. Ibid **55**: 199–207 (13 Figs.).
——— A brief résumé of trematode studies in Washington. The Biolog (Univ. Portland) **3**: 1–2; 9–10.
——— Trematodos ectoparasitos de los peces de las costas del Pacifico. An. Inst. Biol., Univ. Nac. Mexico **7**: 457–467 (7 Figs.).
——— Note on embryo porpoise. Murrelet **17**: 56.
——— Two new ectoparasitic trematodes from the sting-ray, *Myliobatus californicus*. Am. Midland Naturalist **17**: 954–964 (12 Figs.).
1937 Lowell Clyde Lloyd (1903–1936). Tr. Am. Micr. Soc. **56**: 119.
——— (with Kelshaw Bonham) Notes on *Microcotyle sebastis* Goto from Puget Sound. J. Parasitol. **23**: 281–290 (14 Figs.).
——— A method for rearing *Nereis agassizi* and *N. procera*. *In* Galstoff et al: Culture Methods for Invertebrate Animals, Ithaca, N. Y., Comstock Pub. Co., pp. 184–185.

1938 (with Kelshaw Bonham) Ectoparasitic trematodes of Puget Sound fishes—Acanthocotyle. Am. Midland Naturalist **20**: 590–602 (10 Figs.).
1939 When does the robin begin to sing? Murrelet **20**: 18.
1940 (with H. H. Hotson) A fly maggot attacking young birds, with observations on its life history. Ibid **21**: 65–68 (4 Figs.).

PREPARED FOR PUBLICATION

List of animals dredged in Puget Sound.
List of animals dredged off British Columbia and Southeastern Alaska.
The morphology and behavior of *Pteraster tesselatus* Ives. With Irma Rodenhouse)
Further studies on monogenetic trematodes from Puget Sound fish. (With Kelshaw Bonham)
Studies on Hexamita in the Fowl and the Turkey. (With Joseph B. Trainer)